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PHYLOGENY OF BASAL
EUDICOTS BASED ON THREE
MOLECULAR DATA SETS:
atpB, *rbcL*, AND 18S
NUCLEAR RIBOSOMAL DNA
SEQUENCES¹

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ABSTRACT

Phylogenetic analyses were conducted for 73 genera of “lower” eudicots (Ranunculidae and “lower” Hamamelididae), magnoliid outgroups, and appropriate representatives for higher taxa within the “higher” eudicot clade (e.g., Rosidae, Dilleniidae, Asteridae) based on sequences of three genes: the two chloroplast genes *atpB* and *rbcL* and nuclear ribosomal 18S DNA. Based on the partition homogeneity test, the three data sets were relatively congruent ($P \geq 0.13$). The data were analyzed using heuristic parsimony searches and bootstrap analyses in three ways: individually, the two chloroplast sequences combined, and all three sequences combined. Both ingroup and outgroup sampling were varied to test the stability of the tree topology. The trees resulting from a combination of the chloroplast data and all three data sets had the best resolution and the strongest branch support. The following higher taxonomic groups were recognized with high bootstrap values ($> 90\%$): Eudicots (including *Nelumbo*), Ranunculidae (including *Euptelea*), Papaverales, “core” ranunculids, a clade consisting of “lower” hamamelids and “core” eudicots, “core” eudicots (including caryophyllids, asterids, and rosids), Dilleniaceae, caryophyllids (including *Simmondsia*), and asterids. All ranunculid families, including Circaeasteraceae s.l. (including *Kingdonia*) and Lardizabalaceae s.l. (including *Sargentodoxa*), formed well-supported monophyletic groups. Other well-supported eudicot clades were *Platanus*/Proteaceae, Buxaceae/*Didymela*, Trochodendraceae/Tetracentraceae, and a group with poor internal resolution that included genera in Hamamelidaceae, various rosids, and *Paeonia*. Morphology (especially floral features) and other characteristics are described in some detail for well-supported clades determined by the molecular data.

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Recent cladistic analyses of phylogenetic relationships within angiosperms recognize two major clades, eudicots and monocotyledons, both nested within a small assemblage of “basal” dicotyledons at the magnoliid grade (Crane, 1989; Drinnan et al., 1994; APG, 1998). Eudicots are characterized by the possession of triaperturate or triaperturate-derived pollen, and monophyly of the group is further supported by phylogenetic analyses based on morphological and/or molecular data (Chase et al., 1993; Albert et al., 1994; Doyle et al., 1994; Hoot & Crane, 1995; Soltis et al., 1997). Eudicots comprise approximately 75% of extant angiosperm species (subclasses Nelumbonidae, Ranunculidae, Caryophyllidae, Hamamelididae, Dilleniidae, Rosidae, Cornidae, Asteridae, and Lamiidae sensu Takhtajan, 1997). Clarification of phylogenetic patterns at the base of this clade is therefore important to our understanding of relationships among angiosperms as a whole.

Recent discussions have highlighted two assemblages of families as potentially “basal” within the eudicots as a whole: the ranunculids and the “lower” hamamelids (Crane, 1989; Drinnan et al., 1994; Hoot & Crane, 1995). Ranunculidae have sometimes been placed within Magnoliidae (e.g., Ranunculales of Cronquist, 1981), largely on the basis of their frequently polymerous flowers (Drinnan et al., 1994), but are more appropriately included within the eudicots because of their triaperturate pollen (Takhtajan, 1997; Crane, 1989; Drinnan et al., 1994). “Lower” hamamelids, such as *Trochodendron*, *Tetracentron*, and to a lesser extent *Euptelea* and *Cercidiphyllum*, have been thought to retain a variety of unmodified plesiomorphic features from their putative ancestors at the magnoliid grade (cf. Endress, 1986; Crane, 1989).

Because of the extreme diversity in the eudicot clade (ca. 175,000 species), one problem in resolving eudicot phylogeny has been the development of a strategy that provides adequate representation of the group while at the same time allowing sample size to be maintained at a level that is manageable given currently available techniques for phylogenetic analyses. To minimize sampling size, we conducted an extensive survey of phylogenetic patterns within the eudicot clade based on existing molecular data. We then used a “placeholder” strategy to represent the major eudicot groups currently recognized. Also factored into the selection of appropriate taxa were previous phylogenetic analyses of major ranunculid and “lower” hamamelid families, by us and others, which provide acceptable representation of large groups such as the Papaverales,

Ranunculaceae, and Berberidaceae (e.g., Hoot et al., 1995, 1997; Kim & Jansen, 1995).

In this paper, we present the results of phylogenetic analyses of the “lower” eudicots (Ranunculidae and “lower” Hamamelididae) using 73 taxa including appropriate magnoliid outgroups and placeholders for higher taxa within the eudicot clade (e.g., Rosidae, Dilleniidae). Analyses are based on three sequence data sets, both individually and combined: the two chloroplast genes, *atpB* and *rbcL*, and nuclear ribosomal 18S DNA. Because relationships within the magnoliid grade are unresolved (see below), we explore potential changes in tree topology that may result from the use of alternative outgroups. We also explore the effects on tree topology of different sampling strategies within the “lower” eudicot ingroup. Phylogenetic trees resulting from the analyses of molecular data are used to examine evolution of specific morphological, anatomical, and chemical characters.

MATERIALS AND METHODS

TAXON SAMPLING—INGROUP

The 73 taxa included in the analyses presented here were selected to maximize systematic coverage within the Ranunculidae, “lower” Hamamelidae, and other subclasses (Tables 1, 2). Unless otherwise noted, taxonomic groupings are as in Takhtajan (1997; Table 1).

Ranunculidae. Most of the families in the ranunculid complex recognized by various workers (Dahlgren, 1980; Cronquist, 1981; Thorne, 1992; Takhtajan, 1997) were included in our sampling (Table 1). To represent relatively large ranunculid families, placeholders were selected based on previous analyses of molecular and morphological data.

Pteridophyllum, *Hypecoum*, *Dicentra*, and *Corydalis* were chosen as placeholders for Papaverales based on morphological analyses (Kadereit et al., 1995) and combined analyses of molecular and morphological data sets (Hoot et al., 1997). These studies show a sister-group relationship between *Pteridophyllum* and all remaining Papaverales (Fumariaceae and Papaveraceae). The Fumariaceae (represented here by *Hypecoum*, *Dicentra*, and *Corydalis*) are sister to Papaveraceae s. str.

Kingdonia and *Circaeaster*, frequently treated as monotypic families, were included as genera of uncertain affinities within Ranunculidae. Previous analyses have shown that together they form the sister group to a clade composed of *Sargentodoxa* and Lardizabalaceae (Hoot & Crane, 1995).

Table 1. Genera of basal eudicots sampled based on Takhtajan's classification (1997).

Magnoliidae	Lardizabalales	Hamamelidaceae	Ericanae
Magnolianaes	Sargentodoxaceae	<i>Corylopsis</i>	Bruniales
Winterales	<i>Sargentodoxa</i>	<i>Disanthus</i>	Bruniaceae
Winteraceae	Lardizabalaceae	<i>Exbucklandia</i>	<i>Berzella</i>
<i>Drimys</i>	<i>Decaisnea</i>	<i>Hamamelis</i>	Rosidae
<i>Pseudowintera</i>	<i>Sinofranchetia</i>	Altingiaceae	Saxifraganae
<i>Tasmannia</i>	Menispermiales	<i>Altingia</i>	Cunoniales
Illiciales	Menispermaceae	<i>Liquidambar</i>	Eucryphiaceae
Illiciaceae	<i>Menispermum</i>	Daphniphyllanae	<i>Eucryphia</i>
<i>Illicium</i>	<i>Tinospora</i>	Daphniphyllales	Saxifragales
Schisandraceae	Glaucidiales	Daphniphyllaceae	Saxifragaceae
<i>Schisandra</i>	Glaucidiaceae	<i>Daphniphyllum</i>	<i>Heuchera</i>
Austrobaileales	<i>Glaucidium</i>	Buxanae	Iteaceae
Austrobaileyaceae	Hydrastidales	Didymelales	<i>Itea</i>
<i>Austrobaileya</i>	Hydrastidaceae	Didymelaceae	Francoales
Aristolochiales	<i>Hydrastis</i>	<i>Didymeles</i>	Francoaceae
Aristolochiaceae	Berberidales	Buxales	<i>Francoa</i>
<i>Aristolochia</i>	Nandinaceae	Buxaceae	Haloragales
<i>Asarum</i>	<i>Nandina</i>	<i>Buxus</i>	Haloragaceae
Piperanae	Podophyllaceae	<i>Pachysandra</i>	<i>Haloragis</i>
Piperales	<i>Caulophyllum</i>	<i>Styloceras</i>	Gunnerales
Peperomiaceae	<i>Podophyllum</i>	Simmondsiales	Gunneraceae
<i>Peperomia</i>	Ranunculales	Simmondsiaceae	<i>Gunnera</i>
Saururaceae	Ranunculaceae	<i>Simmondsia</i>	Rutanae
<i>Houttuynia</i>	<i>Coptis</i>	Caryophyllidae	Sabiales
<i>Saururus</i>	<i>Xanthorhiza</i>	Caryophyllanae	Sabiaceae
Lauranae	Paeoniales	Caryophyllales	<i>Sabia</i>
Chloranthales	Paeoniaceae	Phytolaccaceae	Coriariales
Chloranthaceae	<i>Paeonia</i>	<i>Phytolacca</i>	Coriariaceae
<i>Chloranthus</i>	Hamamelididae	Molluginaceae	<i>Coriaria</i>
<i>Sarcandra</i>	Trochodendranae	<i>Limeum</i>	Geranianae
Nelumbonidae	Trochodendrales	Chenopodiaceae	Geraniales
Nelumbonales	Trochodendraceae	<i>Spinacia</i>	Geraniaceae
Nelumbonaceae	<i>Trochodendron</i>	Polygonanae	<i>Geranium</i>
<i>Nelumbo</i>	Tetracentraceae	Polygonales	Proteanae
Ranunculidae	<i>Tetracentron</i>	Polygonaceae	Proteales
Ranunculanae	Cercidiphyllales	<i>Rheum</i>	Proteaceae
Papaverales	Cercidiphyllaceae	Dilleniidae	<i>Placospermum</i>
Pteridophyllaceae	<i>Cercidiphyllum</i>	Nepenthanae	<i>Roupala</i>
<i>Pteridophyllum</i>	Eupteleales	Nepenthales	Cornidae
Hypecoaceae	Eupteleaceae	Nepenthaceae	Cornanae
<i>Hypecoum</i>	<i>Euptelea</i>	<i>Nepenthes</i>	Hydrangeales
Fumariaceae	Myrothamnanae	Dilleniaceae	Hydrangeaceae
<i>Corydalis</i>	Myrothamnales	Dilleniales	<i>Hydrangea</i>
<i>Dicentra</i>	Myrothamnaceae	Dilleniaceae	Aralianae
Circaeasterales	<i>Myrothamnus</i>	<i>Dillenia</i>	Araliales
Circaeasteraceae	Hamamelidanae	<i>Hibbertia</i>	Araliaceae
<i>Circaeaster</i>	Hamamelidales	<i>Schumacheria</i>	<i>Hedera</i>
Kingdoniaceae	Platanaceae		
<i>Kingdonia</i>	<i>Platanus</i>		

Sargentodoxa is often assigned to a family of its own, usually considered to be closely related to, or within, Lardizabalaceae (Hoot & Crane, 1995). *Sinofranchetia* and *Decaisnea* were included as placeholders for Lardizabalaceae s. str. based on previous analyses of morphological (Loconte & Estes, 1989; Loconte et al., 1995) and molecular sequence data (Hoot et al., 1995).

We selected *Tinospora* and *Menispermum* as

placeholders for Menispermaceae, based on their diverse fruit and floral morphology (Thanikaimoni, 1984). Preliminary phylogenetic analyses of 17 genera within the family, based on molecular data (*atpB* and *rbcL*), confirmed the monophyly of the family and the relative divergence of *Tinospora* and *Menispermum* (Hoot, unpublished results).

Nandina, *Caulophyllum*, and *Podophyllum* were selected as placeholders for Berberidales based on

Table 2. Species sequenced, voucher information, sources of sequences and accession numbers. KBG = Kirstenbosch Botanical Garden (accession numbers for living plants); SA = Strybing Arboretum.

Family	Species	18S		
		<i>atpB</i> Voucher information/ source of sequence/ GenBank	<i>rbcL</i> Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank
Altingiaceae	<i>Altingia excelsa</i> Noronha	Hoot 9225 (UWM) Hoot AF092103		
Altingiaceae	<i>Altingia</i> sp.		Qiu 93006 (NCU) Qiu AF061996	Qiu 93006 (NCU) Soltis U42552
Altingiaceae	<i>Liquidambar styraciflua</i> L.	Kron 162 (NCU) Hoot AF092104	Bousquet M58394	Soltis & Soltis 2516 (WS) Soltis U42553
Araliaceae	<i>Hedera helix</i> L.	Hoot 972 (UWM) Hoot AF092105	Jansen s.n. (MICH) Olmstead et al. 1992 L01924	Plunkett 1368 (WS) Soltis & Soltis U42500
Aristolochiaceae	<i>Aristolochia macrophylla</i> Lam.		Qiu 91019 (NUC) Qiu L12630	
Aristolochiaceae	<i>A. tomentosa</i> Sims	Nickrent 2922 (SIU) Hoot AF092106		Nickrent 2922 (SIU) Nickrent L24083
Aristolochiaceae	<i>Asarum canadensis</i> L.	Hoot 923 (UWM) Hoot U86383		Nickrent 2888 (SIU) Nickrent L24043
Austrobaileyaaceae	<i>Austrobaileya scandens</i> C. T. White	Qiu 90030 (NCU) Hoot AF092107	Qiu 90030 (NCU) Qiu L12632	Nickrent 2953 (SIU) Nickrent U42503
Berberidaceae	<i>Caulophyllum thalictroides</i> (L.) Michx.	Hoot 925 (UWM) Hoot AF092108	Les s.n. (CONN) Les L08760	Hoot 925 (UWM) Hoot L54064
Berberidaceae	<i>Podophyllum peltatum</i> L.	Hoot 943 (UWM) Hoot AF092109	Hoot 943 (UWM) Hoot AF093716	Nickrent 2891 (SIU) Nickrent L24413

Table 2. Continued.

Family	Species	atpB		rbcL		18S	
		Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank		
Bruniaceae	<i>Berzelia lanuginosa</i> (L.) Brongn.	KBG 7589	Price s.n. (IND)	Price s.n. (IND)	Price s.n. (IND)		
		Savolainen AF095731	Price & Rodman L14391	Soltis U42508			
Buxaceae	<i>Buxus sempervirens</i> L.	Hoot 921 (UWM)	Hoot 921 (UWM)	Hoot 921 (UWM)			
		Hoot AF092110	Hoot AF093717	Hoot L54065			
Buxaceae	<i>Pachysandra procumbens</i> Michx.	Hoot 917 (UWM)	Hoot 917 (UWM)	Hoot 917 (UWM)			
		Hoot AF092111	Hoot AF093718	Hoot AF094533			
Buxaceae	<i>Styloceras laurifolium</i> (Willd.) Kunth	Dillon et al. 6398 (F)	Dillon et al. 6398 (F)	Dillon et al. 6398 (F)			
Cercidiphyllaceae	<i>Cercidiphyllum japonicum</i> Siebold & Zucc.	Hoot AF093421	Hoot AF093733	Hoot AF094563			
		Hoot AF092112	Olmstead 90-016 (COLO)	Olmstead 90-016 (COLO)			
Chenopodiaceae	<i>Spinacia oleracea</i> L.	Zurawski U23082	Zurawski V00168	Nickrent 2896 (SIU)			
			Chase 204 (NCU) Hills L12640	Nickrent L24420			
Chloranthaceae	<i>Chloranthus japonicus</i> Siebold						
Chloranthaceae	<i>C. oldhami</i> Solms	Lammers 8575 (F)		Lammers 8575 (F)			
		Hoot AF092113		Hoot AF094535			
Chloranthaceae	<i>Sarcandra glabra</i> (Thunb.) Nakai	Lammers 8542 (F)		Lammers 8542 (F)			
		Hoot AF092114		Hoot AF094536			
Chloranthaceae	<i>S. grandifolia</i> (Miq.) Subr. & A. N. Henry		Qiu 92002 (NCU)				
			Qiu L12663				

Family	Species	<i>atpB</i> Voucher information/ source of sequence/ GenBank	<i>rbcL</i> Voucher information/ source of sequence/ GenBank	18S Voucher information/ source of sequence/ GenBank
Circaeasteraceae	<i>Circaeaster agrestis</i> Maxim.	Chase 506 (K) Hoot AF092116	Chase 506 (K) Hoot AF0937120	Chase 506 (K) Hoot AF094538
Coriariaceae	<i>Coriaria myrtifolia</i> L.	Chase 245 (NCU) Hoot AF092117	Chase 245 (NCU) Hoot L01897	Chase 245 (NCU) Hoot AF094539
Daphniphyllaceae	<i>Daphniphyllum</i> sp.	Wagner <i>et al.</i> 6599 (HAST) Hoot AF092118	Qiu 91026 (NCU) Qiu L01901	Wagner <i>et al.</i> 6599 (HAST) Hoot AF094540
Didymelaceae	<i>Didymeles</i> sp.	<i>Andrianantonnina</i> 387 (MO) Hoot AF092119	<i>Andrianantonnina</i> 387 (MO) Chase AF061994	<i>Andrianantonnina</i> 387 (MO) Hoot AF094541
Dilleniaceae	<i>Dillenia alata</i> (DC.) Mart.		Chase 234 (NCU) Kron L01903	<i>Nickrent</i> 2956 (SIU) Nickrent U38314
Dilleniaceae	<i>D. retusa</i> Thunb.	Chase 2103 (K) Savolainen & Morton AF095732		
Dilleniaceae	<i>Hibbertia volubilis</i> Andr.	Hoot 9222 (UWM) Hoot AF092120	Hoot 9222 (UWM) Hoot AF093721	Hoot 9222 (UWM) Hoot AF094542
Dilleniaceae	<i>Schumacheria</i> sp.	Chase 308 (K) Hoot AF092121	Chase 308 (K) Hoot AF095734	Chase 308 (K) Hoot AF094543
Eucryphiaceae	<i>Eucryphia cordifolia</i> Cav.	Hoot 9224 (UWM) Hoot AF092122		
Eucryphiaceae	<i>E. lucida</i> Druce		SA 86-0250 Morgan & Soltis L01918	SA 86-0250 Soltis U42533

Table 2. Continued.

Family	Species	18S		
		<i>atpB</i> Voucher information/ source of sequence/ GenBank	<i>rbcL</i> Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank
Eupteleaceae	<i>Euptelea polyandra</i> Siebold & Zucc.	<i>Qiu 90026</i> (NCU)	<i>Qiu 90026</i> (NCU)	<i>Qiu 90026</i> (NCU)
		Hoot U86384	<i>Qiu</i> L12645	Hoot L75831
Francoaceae	<i>Francoa appendiculata</i> A. Juss.	<i>Chase 2502</i> (K) Morton & Savolainen AF035905		
Francoaceae	<i>F. sonchifolia</i> Cav.		<i>Soltis & Soltis 2479</i> (WS)	<i>Soltis & Soltis 2479</i> (WS)
Fumariaceae	<i>Corydalis nobilis</i> Pers.	<i>Hoot 9225</i> (UWM)	<i>Hoot 9225</i> (UWM)	<i>Hoot 9225</i> (UWM)
		Hoot AF093372	Hoot AF093722	Hoot AF094544
Fumariaceae	<i>Dicentra eximia</i> Torr.	<i>Reznicek 9756</i> (MICH)	<i>Reznicek 9756</i> (MICH)	<i>Reznicek 9756</i> (MICH)
Geraniaceae	<i>Geranium cinereum</i> Cav.	Hoot L37927	Hoot L37917	Hoot L37908
		<i>Hoot 971</i> (UWM)	<i>Price s.n.</i> (IND)	<i>Price s.n.</i> (IND)
Glaucidiaceae	<i>Glaucidium palmatum</i> Siebold & Zucc.	Hoot AF093373	<i>Price</i> L14695	Soltis U42541
		<i>Hoot 924</i> (UWM)	<i>Hoot 924</i> (UWM)	<i>Hoot 924</i> (UWM)
Gunneraceae	<i>Gunnera hamiltonii</i> Kirk ex W. S. Ham.	Hoot AF093375	Hoot L75848	Hoot L75829
		<i>Chase 562</i> (K)	<i>Chase 562</i> (K)	<i>Chase 562</i> (K)
Haloragaceae	<i>Haloragis erecta</i> Eichler	Hoot AF093374	Hoot AF093724	Hoot AF094546
		<i>Chase 594</i> (K)	<i>Chase 594</i> (K)	<i>Chase 594</i> (K)
Haloragaceae	<i>H. serra</i> Brongn.	Hoot AF093376		Hoot AF094547
Haloragaceae	<i>H. serra</i> Brongn.		<i>Conti 195</i> (WIS)	
			Conti U26325	

Table 2. Continued.

Family	Species	atpB			rbcL			18S		
		Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	
Hamamelidaceae	<i>Corylopsis pauciflora</i> Siebold & Zucc.	Reznicek 9239 (MICH) Hoot AF093377	Reznicek 9239 (MICH) Hoot AF060710	Reznicek 9239 (MICH) Hoot AF060710	Reznicek 9239 (MICH) Hoot AF094548	Reznicek 9239 (MICH) Hoot AF094548				
Hamamelidaceae	<i>Disanthus cercidifolius</i> Maxim.	Hoot 9221 (UWM) Hoot AF093378	Hoot 9221 (UWM) Hoot AF060709	Hoot 9221 (UWM) Hoot AF060709	Hoot 9221 (UWM) Hoot AF094549	Hoot 9221 (UWM) Hoot AF094549				
Hamamelidaceae	<i>Exbucklandia populnea</i> R. Br. ex Griff.	Chase 619 (K) Hoot AF093379	Chase 619 (K) Hoot AF060708	Chase 619 (K) Hoot AF060708	Chase 619 (K) Hoot AF094550	Chase 619 (K) Hoot AF094550				
Hamamelidaceae	<i>Hamamelis mollis</i> Oliv.		Qiu 91035 (NCU) Qiu L01922	Qiu 91035 (NCU) Qiu L01922						
Hamamelidaceae	<i>H. virginiana</i> L.	Hoot 910 (UWM) Hoot AF093380			Hoot 910 (UWM) Hoot AF094551	Hoot 910 (UWM) Hoot AF094551				
Hydrangeaceae	<i>Hydrangea arborescens</i> L.	Hoot 971 (UWM) Hoot AF093381								
Hydrangeaceae	<i>H. macrophylla</i> Torr.		Morgan 2150 (WS) Soltis L11187	Morgan 2150 (WS) Soltis L11187	Morgan 2150 (WS) Soltis U42781	Morgan 2150 (WS) Soltis U42781				
Hydrastidaceae	<i>Hydrastis canadensis</i> L.	Naczi 2883 (MICH) Hoot AF093382	Naczi 2883 (MICH) Hoot L75849	Naczi 2883 (MICH) Hoot L75849	Naczi 2883 (MICH) Hoot L75828	Naczi 2883 (MICH) Hoot L75828				
Hypecoaceae	<i>Hypecoum imberbe</i> Sm.	Chase 528 (K) Hoot U86398	Chase 528 (K) Hoot U86628	Chase 528 (K) Hoot U86628	Chase 528 (K) Hoot AF094553	Chase 528 (K) Hoot AF094553				
Illiciaceae	<i>Illicium parviflorum</i> Michx. ex Vent.	Naczi 2784 (MICH) Hoot U86385	Qiu 83 (NCU) Qiu L12652	Qiu 83 (NCU) Qiu L12652	Naczi 2784 (MICH) Hoot L75832	Naczi 2784 (MICH) Hoot L75832				

Table 2. Continued.

Family	Species	atpB			rbcL			18S		
		Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	
Iteaceae	<i>Itea ilicifolia</i> Oliv.	MacDougal 5060 (MO) Hoot AF093383			Ware 9401 (WS) Soltis L11188			Ware 9401 (WS) Soltis U42545		
Iteaceae	<i>I. virginica</i> L.									
Kingdoniaceae	<i>Kingdonia uniflora</i> Balf. f. & W. W. Sm.	Qin s.n. (PE) Hoot AF092115			Qin s.n. (PE) Hoot AF093719			Qin s.n. (PE) Hoot AF094537		
Lardizabalaceae	<i>Decaisnea fargesii</i> Franch.	Reznicek 9236 (MICH) Hoot L37926			Reznicek 9236 (MICH) Hoot L37907			Reznicek 9236 (MICH) Hoot L37916		
Lardizabalaceae	<i>Sinofranchetia chinensis</i> Helmsl.	Hoot 9220 (UWM) Hoot/Culham L37931			Hoot 9220 (UWM) Hoot L37912			Hoot 9220 (UWM) Hoot L37921		
Menispermaceae	<i>Menispermum canadensis</i> L.	Naczi 2837 (MICH) Hoot AF093384			Naczi 2837 (MICH) Hoot AF093726			Naczi 2837 (MICH) Hoot L75834		
Menispermaceae	<i>Tinospora caffra</i> Miers	Jaarsveld 2131 (NBG) Hoot L37933			Jaarsveld 2131 (NBG) Hoot L37923			Jaarsveld 2131 (NBG) Hoot L37914		
Molluginaceae	<i>Limeum</i> sp.	Hoot 983 (UWM) Hoot AF093385			Hoot 983 (UWM) Hoot AF093727			Hoot 983 (UWM) Hoot AF094554		
Myrothamnaceae	<i>Myrothamnus flabellifolius</i> Welw.	Winter 72 (RAV) Hoot AF093386			Winter 72 (RAV) Hoot AF060707			Winter 72 (RAV) Hoot AF094555		
Nandinaeae	<i>Nandina domestica</i> Thunb.	Hoot 922 (UWM) Hoot L37930			Hoot 922 (UWM) Hoot L37920			Hoot 922 (UWM) Hoot L37911		

Table 2. Continued.

Family	Species	18S		
		<i>atpB</i> Voucher information/ source of sequence/ GenBank	<i>rbcl</i> Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank
Nelumbonaceae	<i>Nelumbo lutea</i> (Willd.) Pers.	Hoot 974 (UWM)	<i>Les s.n.</i> (CONN)	Hoot 974 (UWM)
		Hoot AF093387	Les/Qiu M77032	Hoot AF094556
Nepenthaceae	<i>Nepenthes alata</i> Blanco	Chase 145 (NCU)	Chase 145 (NCU)	
		Hoot AF093388	Chase L01935	
Nepenthaceae	<i>Nepenthes</i> sp.			Nickrent 3056 (SIU)
Paeoniaceae	<i>Paeonia suffruticosa</i> Andrews	Reznicek 9235 (MICH)		Soltis U42787
		Hoot AF093389		Chase 486 (K) Soltis U42792
Paeoniaceae	<i>P. tenuifolia</i> L.		Kron 2115 (NCU)	
Peperomiaceae	<i>Peperomia serpens</i> (Sw.) Loudon	DN 2907 (SIU)	Kron L13187	DN 2907 (SIU)
		Hoot AF093390		Nickrent L24411
Peperomiaceae	<i>Peperomia</i> sp.		Qiu 91047 (NCU)	
			Qiu L12661	
Phytolaccaceae	<i>Phytolacca americana</i> L.	Hoot 942 (UWM)	Rettig 1651 (TAMU)	Hoot 942 (UWM)
		Hoot AF093391	Rettig et al. M62567	Hoot AF094557
Platanaceae	<i>Platanus occidentalis</i> L.	Qiu P90005 (NCU)	Qiu P90005 (NCU)	Soltis & Soltis 2514 (WS)
		Hoot U86386	Qiu L01943	Sweere, Zimmer & Soltis U42794
Polygonaceae	<i>Rheum rhaponticum</i> L.	Hoot 953 (UWM)		Hoot 953 (UWM)
		Hoot AF093392	Giannassi et al. M77702	Hoot AF094558

Table 2. Continued.

Family	Species	atpB			rbcL			18S		
		Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	
Proteaceae	<i>Placospermum coriaceum</i> C. T. White & W. D. Francis	Douglas 110 (MEL) Hoot AF060391	Douglas 110 (MEL) Hoot AF093729	Douglas 110 (MEL) Hoot L75837	Douglas 131 (MEL) Hoot AF094559	Douglas 131 (MEL) Hoot AF094559	Douglas 131 (MEL) Hoot AF094559	Douglas 131 (MEL) Hoot AF094559	Douglas 131 (MEL) Hoot AF094559	
Proteaceae	<i>Roupala macrophylla</i> Pohl	Douglas 131 (MEL) Hoot AF060416	Douglas 131 (MEL) Hoot AF093728	Douglas 131 (MEL) Hoot AF093728	Chase 531 (K) Hoot U86631	Chase 531 (K) Hoot U86631	Chase 531 (K) Hoot U86631	Chase 531 (K) Hoot AF094560	Chase 531 (K) Hoot AF094560	
Pteridophyllaceae	<i>Pteridophyllum racemosum</i> Siebold & Zucc.	Chase 531 (K) Hoot U86400	Chase 531 (K) Hoot U86631	Chase 531 (K) Hoot U86631	Voss & Howard s.n. (MICH) Hoot AF093393	Voss & Howard s.n. (MICH) Hoot AF093393	Voss & Howard s.n. (MICH) Hoot AF093393	Voss & Howard s.n. (MICH) Hoot AF093393	Voss & Howard s.n. (MICH) Hoot AF093393	
Ranunculaceae	<i>Coptis trifolia</i> (L.) Salisb.	Voss & Howard s.n. (MICH) Hoot AF093393	Voss & Howard s.n. (MICH) Hoot AF093393	Voss & Howard s.n. (MICH) Hoot AF093393	Voss & Howard s.n. (MICH) Hoot AF093393	Voss & Howard s.n. (MICH) Hoot AF093393	Voss & Howard s.n. (MICH) Hoot AF093393	Voss & Howard s.n. (MICH) Hoot AF093393	Voss & Howard s.n. (MICH) Hoot AF093393	
Ranunculaceae	<i>Xanthorhiza simplicissima</i> Marshall	Qiu 91030 (NCU) Hoot AF093394	Qiu 91030 (NCU) Qiu L12669	Qiu 91030 (NCU) Hoot AF093394	Qiu 91030 (NCU) Qiu L12669	Qiu 91030 (NCU) Qiu L12669	Qiu 91030 (NCU) Hoot L75839	Qiu 91030 (NCU) Hoot L75839	Qiu 91030 (NCU) Hoot L75839	
Sabiaceae	<i>Sabia swinhoei</i> Hemsl. ex F. B. Forbes & Hemsl.	Wagner 6158 (HAST) Hoot AF093395	Wagner 6158 (HAST) Hoot AF093395	Wagner 6158 (HAST) Hoot AF093395	Wagner 6158 (HAST) Hoot AF093395	Wagner 6158 (HAST) Hoot AF093395	Wagner 6158 (HAST) Hoot AF093395	Wagner 6158 (HAST) Hoot AF093395	Wagner 6158 (HAST) Hoot AF093395	
Sabiaceae	<i>Sabia</i> sp.									
Sargentodoxaceae	<i>Sargentodoxa cuneata</i> (Oliv.) Rehder & E. H. Wilson	Qin s.n. (PE) Hoot AF093396	Qin s.n. (PE) Hoot AF093731	Qin s.n. (PE) Hoot AF093396	Qin s.n. (PE) Hoot AF093731	Qin s.n. (PE) Hoot AF093731	Qin s.n. (PE) Hoot L75841	Qin s.n. (PE) Hoot L75841	Qin s.n. (PE) Hoot L75841	
Saururaceae	<i>Houttuynia cordata</i> Thunb.	Reznicek 9238 (MICH) Hoot AF093397	Les s.n. (CONN) Les L08762	Reznicek 9238 (MICH) Hoot AF093397	Les s.n. (CONN) Les L08762	Les s.n. (CONN) Les L08762	Nickrent 2940 (SIU) Nickrent L24147	Nickrent 2940 (SIU) Nickrent L24147	Nickrent 2940 (SIU) Nickrent L24147	
Saururaceae	<i>Saururus cernuus</i> L.	Olmstead 88-006 (COLO) Hoot AF093398	Olmstead 88-006 (COLO) Olmstead L14294	Olmstead 88-006 (COLO) Hoot AF093398	Olmstead 88-006 (COLO) Olmstead L14294	Olmstead 88-006 (COLO) Olmstead L14294	Suh 128 (US) Sweere, Zimmer & Soltis U42805	Suh 128 (US) Sweere, Zimmer & Soltis U42805	Suh 128 (US) Sweere, Zimmer & Soltis U42805	

Family	Species	atpB			rbcL			18S		
		Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	Voucher information/ source of sequence/ GenBank	
Saxifragaceae	<i>Heuchera micrantha</i> Douglas		Soltis & Soltis 1949 (WS) Soltis & Soltis L01925	Soltis & Soltis 1949 (WS) Soltis & Soltis X28139						
Saxifragaceae	<i>H. sanguinea</i> Engelm.	Hoot 932 (UWM) Hoot AF093399								
Schisandraceae	<i>Schisandra chinensis</i> (Turcz.) Baill.	Reznicek 10720 (MICH) Hoot AF093400		Reznicek 10720 (MICH) Hoot AF094561						
Schisandraceae	<i>S. sphenanthera</i> Rehder & E. H. Wilson		Qiu 73 (NCU) Qiu L12665							
Simmondsiaceae	<i>Simmondsia chinensis</i> C. K. Schneid.	S. Boyd et al. 3355 (F) Hoot AF093401	S. Boyd et al. 3355 (F) Hoot AF093732	S. Boyd et al. 3355 (F) Hoot AF094562						
Tetracentraceae	<i>Tetracentron sinensis</i> Oliv.	Qiu 90009 (NCU) Hoot AF093422	Qiu 90009 (NCU) Qiu L12668	Qiu 90009 (NCU) Hoot AF094564						
Trochodendraceae	<i>Trochodendron aralioides</i> Siebold & Zucc.	Qiu 90026 (NC) Hoot AF093423	Qiu 90026 (NC) Qiu L01958	Qiu 90026 (NC) Hoot AF094565						
Winteraceae	<i>Drimys winteri</i> J. R. Forst. & G. Forster	Nickrent 3013 (SIU) Hoot AF093425	Nickrent 3013 (SIU) Hoot AF093734	Suh 47 (US) Sweere, Zimmer & Soltis U42823						
Winteraceae	<i>Pseudowintera colorata</i> Dandy	Hoot 9223 (UWM) Hoot AF093426	Hoot 9223 (UWM) Hoot AF093735	Hoot 9223 (UWM) Hoot AF094567						
Winteraceae	<i>Tasmannia insipida</i> R. Br. ex DC.		A. N. Rodd 5540 (NSW) Qiu L01957							
Winteraceae	<i>T. lanceolata</i> (Poir.) A. C. Smith	Raleigh 109 (MEL) Hoot AF093424		Raleigh 109 (MEL) Hoot AF094566						

the frequent separation of *Nandina* as Nandinaeae (e.g., Dahlgren, 1980; Takhtajan, 1997) and the position of *Caulophyllum* and *Podophyllum* close to the base of the Berberidales (Kim & Jansen, 1995; Nickol, 1995).

Glaucidium, *Hydrastis*, *Coptis*, and *Xanthorrhiza* were selected as representatives of Ranunculaceae s.l. based on extensive morphological and molecular phylogenetic studies of the family, which place these genera close to the base of the family (Hoot, 1991, 1995; Johansson & Jansen, 1993; Johansson, 1995).

“Lower” Hamamelididae. The “lower” hamamelids have been only loosely defined by previous workers, but are generally considered to include the orders Trochodendrales (Trochodendraceae, Tetracentraceae), Cercidiphyllales (Cercidiphyllaceae), Eupteleales (Eupteleaceae), Myrothamnales (Myrothamnaceae), Hamamelidales (Platanaceae, Hamamelidaceae, Altingiaceae), Daphniphyllales (Daphniphyllaceae), Didymelales (Didymelaceae), Buxales (Buxaceae), and Simmondsiales (Simmondsiaceae), all belonging to subclass Hamamelididae sensu Takhtajan (1997; Table 1). With the exception of Hamamelidaceae, Altingiaceae, and Buxaceae, each of the families included in these orders contains a single genus, and all were included in our analyses (Table 1).

Hamamelidaceae (sensu Endress, 1989a) include 30 genera distributed among four subfamilies, of which Hamamelidoideae is the largest with 22 genera (Endress, 1989a, 1993). Because results of previous phylogenetic analyses suggest that Hamamelidaceae are not monophyletic (e.g., Morgan & Soltis, 1993; Manos et al., 1993; Chase et al., 1993; Qiu et al., 1998), three of the four subfamilies, i.e., Hamamelidoideae, Exbucklandioideae, and Altingioideae, were represented in our study. A more detailed assessment of the monophyly of Hamamelidaceae and the relationships among the genera will require more extensive sampling within the family.

Several taxa that have been identified as putatively closely related to Hamamelidaceae (sensu Endress, 1989a) based on recent phylogenetic analyses of molecular data were also included in our sampling (e.g., Chase et al., 1993; Morgan & Soltis, 1993; Drinnan et al., 1994; Soltis et al., 1997). In addition to *Cercidiphyllum* and *Daphniphyllum*, these taxa are Saxifragaceae s. str. (equivalent to the Saxifragoideae of Engler, 1930, and Schulze-Menz, 1964, and represented here by *Heuchera*; Soltis et al., 1993; Soltis & Soltis, 1997), other genera of saxifragoid affinity (represented here by *Itea*),

Haloragaceae (represented here by *Haloragis*), and the isolated genus *Paeonia* (placed in Ranunculidae by Takhtajan, 1997).

Preliminary analyses of molecular data for five genera and nine species of Buxales (including *Styloceras*) indicated three genera are appropriate as placeholders for the family: *Styloceras*, *Buxus*, and *Pachysandra* (Hoot, unpublished data). Cronquist (1981) tentatively included Buxaceae in Euphorbiales. However, Takhtajan's (1997) superorder Buxanae, which includes Didymelales, Buxales, and Simmondsiales, is placed in subclass Hamamelididae. The genus *Simmondsia*, which was separated by Cronquist (1981) and Takhtajan (1997) from Buxaceae and placed within a monogeneric family, was also included in our sampling.

Taxa that have been identified by previous phylogenetic analyses (e.g., Chase et al., 1993; Williams et al., 1994; Drinnan et al., 1994; Soltis et al., 1997) as putatively related to “lower” hamamelids were also included in the taxonomic sampling. These include *Nelumbo* (Nelumbonaceae), *Roupala* and *Placospermum* (Proteaceae), *Sabia* (Sabiaceae), and *Gunnera* (Gunneraceae).

Core eudicots. Results of recent phylogenetic analyses (Olmstead et al., 1992; Chase et al., 1993; Drinnan et al., 1994; Williams et al., 1994; Soltis et al., 1997) converge in identifying a large clade, which we term here as “core” eudicots, that includes the majority of eudicot species diversity. Based on recent results derived mainly from analyses of molecular data, the core eudicots are largely distributed in three distinct clades: the “caryophyllids,” “asterids,” and “rosids” (Chase et al., 1993; Soltis et al., 1997), all of which were represented in our sampling.

The “caryophyllid clade,” equivalent to the “caryophyllids” of Search II from Chase et al. (1993) or the Caryophyllidae s.l. of Soltis et al. (1997), was represented in our analyses by genera belonging to five families: Polygonaceae (*Rheum*), Chenopodiaceae (*Spinacia*), Molluginaceae (*Limeum*), Phytolaccaceae (*Phytolacca*), and Nepenthaceae (*Nepenthes*). Nepenthaceae have been recognized recently as closely related to Caryophyllidae (Albert et al., 1992; Chase et al., 1993; Williams et al., 1994; Soltis et al., 1997). Because *Dillenia* appears as either sister to a largely caryophyllid clade (Chase et al., 1993) or closely associated with several “lower” hamamelid species (Qiu et al., 1998), three representatives of the Dilleniaceae were included in our sampling: *Dillenia*, *Hibbertia*, and *Schumacheria*.

The “rosid clade,” corresponding to the “rosids”

of Search II in Chase et al. (1993) and roughly to the Rosidae of Soltis et al. (1997), was represented in our analyses by four placeholders: *Francoa* and *Geranium* (Rosid II), and *Coriaria* and *Eucryphia* (Rosid I). Genera of Hamamelidaceae, *Cercidiphyllum*, *Daphniphyllum*, and other taxa have been identified as forming a paraphyletic grade at the base of the rosid clade (e.g., Morgan & Soltis, 1993; Soltis et al., 1993; Williams et al., 1994) or have been included within the most basal group (i.e., "Rosid III" of Chase et al., 1993, or "Saxifragoids" of Soltis et al., 1997) of the rosid clade (e.g., Chase et al., 1993; Soltis et al., 1997; Soltis & Soltis, 1997).

The "asterid clade," equivalent to the "asterids" of Search II of Chase et al. (1993) and to Asteridae s.l. of Olmstead et al. (1993), was represented in our analyses by three placeholders. The "Cornales" of Olmstead (1993) or "asterid IV" clade of Chase et al. (1993) was represented by *Hydrangea* (Hydrangeaceae). The "asterid II" clade of Chase et al. (1993), which includes the Apiales, Dipsacales, Asterales s.l., and several genera of varied taxonomic affinity (Olmstead et al., 1992; Olmstead et al., 1993; Cosner et al., 1994; Plunkett et al., 1996), was represented by *Hedera* (Araliaceae) and *Berzelia* (Bruniaceae).

TAXON SAMPLING—OUTGROUP

The sister-group relationships of the eudicots as a whole with respect to groups at the magnoliid grade are not resolved, and many different phylogenetic arrangements have been suggested (e.g., Donoghue & Doyle, 1989a; Chase et al., 1993; Doyle et al., 1994; Soltis et al., 1997). Because of these uncertainties and the absence of critical eudicot features in potential outgroup taxa, rooting the basal eudicots is problematic (Donoghue & Doyle, 1989b; Doyle & Donoghue, 1993). Therefore, we selected a broad taxonomic sample of potential woody and herbaceous outgroup taxa within subclass Magnoliidae, representing 13 genera in eight families within six different orders (all sensu Takhtajan, 1997): Winterales (Winteraceae), Illiciales (Illiciaceae, Schisandraceae), Austrobaileyales (Austrobaileyaceae), Aristolochiales (Aristolochiaceae), Piperales (Peperomiaceae, Saururaceae), and Chloranthales (Chloranthaceae; Table 1). For the analyses presented here, *Austrobaileya* was designated as outgroup, allowing for an additional check of eudicot monophyly (Figs. 1–6). In addition, alternative outgroup configurations were chosen to check the effect of outgroup sampling on the tree topologies (described below).

MOLECULAR METHODS

Total cellular DNA was isolated from fresh, herbarium, or silica-dried material using the miniprep method of Doyle and Doyle (1987). The amplification primers and polymerase chain reaction (PCR) protocol are as described in Hoot et al. (1995). Purification of PCR product and manual double-stranded sequencing protocols are as described in Hoot (1995). Automated sequencing from purified PCR product was performed on an ABI automated sequencer (Applied Biosystems, Model 373A) and contigs were assembled using Sequencher[™] 3.0 (Gene Codes Corporation).

Sequence comparisons for *atpB*, *rbcL*, and 18S included 1493, 1397, and 1635 bp, respectively. Both strands of DNA were sequenced for all regions with approximately 80% overlap. Several regions were removed from the 18S data set due to alignment and/or compression problems at the following positions in relation to the soybean 18S sequence (Eckenrode et al., 1985): 131–133, 224–231, 666–669, 1363–1366, and 1512–1517. In addition, three informative sites were removed due to compensatory changes (paired sites indicated in parentheses): 734 (708), 1050 (1076), and 1074 (1052). See Hoot (1995) for details of data collection.

DATA ANALYSIS

Phylogenetic analyses with the complete sampling were performed separately on the *rbcL*, *atpB*, and 18S data as well as on the combined data sets *atpB/rbcL* and *atpB/rbcL/18S*. Analyses were performed with PAUP* 4.0d64 (kindly provided by D. Swofford) using the heuristic search option with 20 random additions, TBR (tree bisection-reconnection branch swapping), and MULPARS (retention of all equally parsimonious trees) in effect. Bootstrap analyses (Felsenstein, 1985) with 100 replications were performed on each individual and combined data set using the heuristic search option with simple addition sequence. In the case of individual data sets (*atpB*, *rbcL*, and 18S), the maxtree limit of 2000 was reached on some bootstrap replications. Alternative tree topologies and resultant changes in tree length were explored using MacClade 3.0 (Maddison & Maddison, 1992).

Alternative analyses. Several additional analyses of the combined data sets were performed as described above to check the effect of taxon sampling on the tree topology. Five alternative samplings were examined as follows: (1) exclusion of paleoherbs (*Chloranthus*, *Sarcandra*, *Asarum*, *Aristolochia*, *Peperomia*, *Saururus*, and *Houttuynia*;

Table 3. Comparison of indices for the various trees illustrated in Figures 1–5. Tree length includes uninformative characters; consistency index (CI) excludes uninformative characters. RI = retention index, RC = rescaled consistency index.

Data set	No. variable characters	No. informative characters	No. of trees	Length of trees	CI	RI	RC
<i>atpB</i>	576	435	108	2233	0.34	0.59	0.23
<i>rbcL</i>	630	431	2	2478	0.31	0.54	0.20
<i>atpB/rbcL</i>	1206	866	6	4744	0.32	0.56	0.21
18S nrDNA	307	175	270	939	0.33	0.58	0.25
<i>atpB/rbcL/18S</i>	1513	1041	15	5714	0.32	0.56	0.21

designated outgroup *Austrobaileya*); (2) exclusion of “woody” magnoliids (*Austrobaileya*, *Illicium*, *Schisandra*, *Pseudowintera*, *Tasmannia*, and *Drimys*; designated outgroup *Chloranthus*); (3) ranunculids reduced to six placeholders (*Euptelea*, *Pteridophyllum*, *Sargentodoxa*, *Menispermum*, *Nandina*, *Hydrastis*); (4) ranunculids reduced to three placeholders (*Euptelea*, *Pteridophyllum*, *Hypocoum*); and (5) separate analysis of “core” eudicots (see Fig. 6) using *Tetracentron* and *Trochodendron* as outgroups.

Congruence of data sets. Before combining the data sets, data (or character) congruence was assessed using the partition homogeneity test (Farris et al., 1995; implemented with PAUP* vers. 4.0), a bootstrap approach that randomly partitions characters. It tests the null hypothesis that a given partition of a data set (for example, *rbcL* and *atpB* data sets) represents a random partition of the data. All combinations of data sets were analyzed with 100 replicates, the heuristic search option with simple addition sequence, TBR, and MULPARS in effect. To reduce computation time, the analyses were done with reduced sampling (50 taxa). The following outgroup and placeholder taxa were omitted from the analyses: *Schisandra*, *Chloranthus*, *Sarcandra*, *Pseudowintera*, *Tasmannia*, *Drimys*, *Asarum*, *Aristolochia*, *Peperomia*, *Saururus*, *Houttuynia*, *Berzelia*, *Hedera*, *Coriaria*, *Francoa*, *Dillenia*,

Schumacheria, *Nepenthes*, *Spinacia*, *Limeum*, *Phytolacca*, *Haloragis*, and *Itea*. To test the effect of the reduced sampling, a heuristic parsimony search of the combined data with reduced sampling was performed. It resulted in a strict consensus tree (of six trees) that was virtually identical in topology to that found in the strict consensus tree with complete sampling (Fig. 6).

RESULTS

The number of variable and potentially phylogenetically informative characters found in each data set, the number and length of most parsimonious trees obtained, and the consistency, retention, and rescaled consistency indices of the various analyses performed are presented in Table 3. The results from the incongruence tests of the reduced data sets are found in Table 4. The *P*-values resulting from the partition homogeneity test indicate that the data partitions are random (*P*-values ranges between 0.13 and 0.41) and that the data sets are reasonably congruent.

One of the most parsimonious trees produced from each analysis is presented to illustrate the support at different nodes (Figs. 1–5). In the results and discussion presented here, clades with bootstrap values of 70% or more are regarded, provisionally, as “well supported” (Hillis & Bull, 1993). Tables 5 and 6 present the bootstrap support for various systematic groupings.

RESULTS BASED ON THE CHLOROPLAST GENES

The two chloroplast sequences, *atpB* and *rbcL*, are most congruent (*P*-value = 0.41, Table 4). Comparison of the strict consensus trees derived from each data set (Figs. 1, 2) indicates minor differences in the degree of resolution of some clades and some positional differences in branches with weak bootstrap support (< 70%). Because the data are congruent and topologically similar, only the

Table 4. *P*-values from partition-homogeneity test, with 100 replications for various partitions of data. To reduce computation time, analyses were done with reduced sampling (see “Materials and Methods”). *P*-values of 0.05 or more indicate that the partition of data sets is random, indicating congruence between data sets.

Data sets	<i>P</i> -value
<i>atpB</i> vs. <i>rbcL</i>	0.41
<i>atpB</i> vs. 18S	0.13
<i>rbcL</i> vs. 18S	0.26
<i>atpB/rbcL</i> vs. 18S	0.21

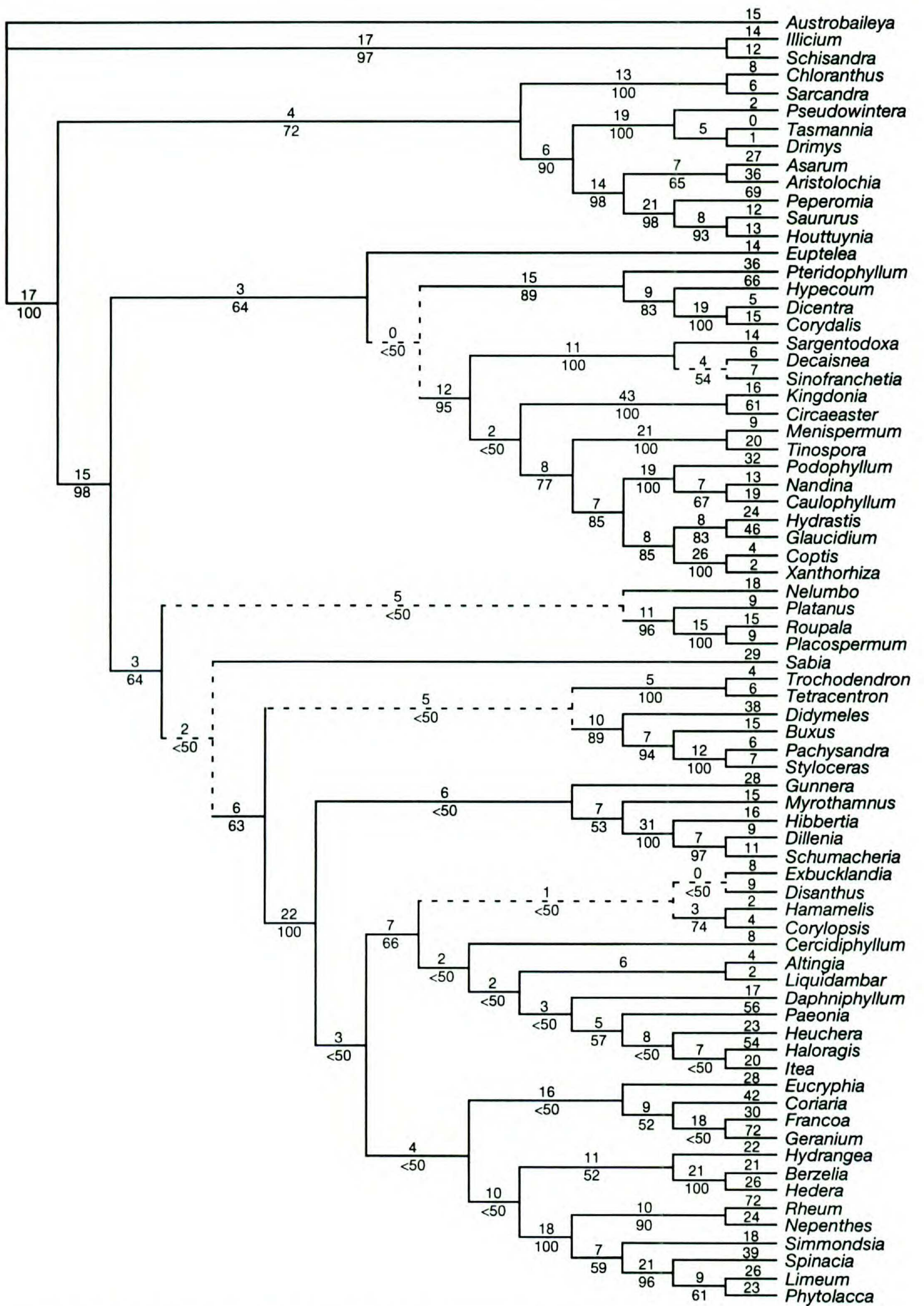


Figure 1. One of 108 shortest trees based on the *atpB* sequence data. Numbers above the line indicate the number of nucleotide changes supporting each branch. Numbers below the branches are the percentage of times that the branch was recovered in 100 bootstrap replications. Dotted lines indicate branches that collapse in the strict consensus tree derived from multiple shortest trees. See Table 3 for tree statistics.

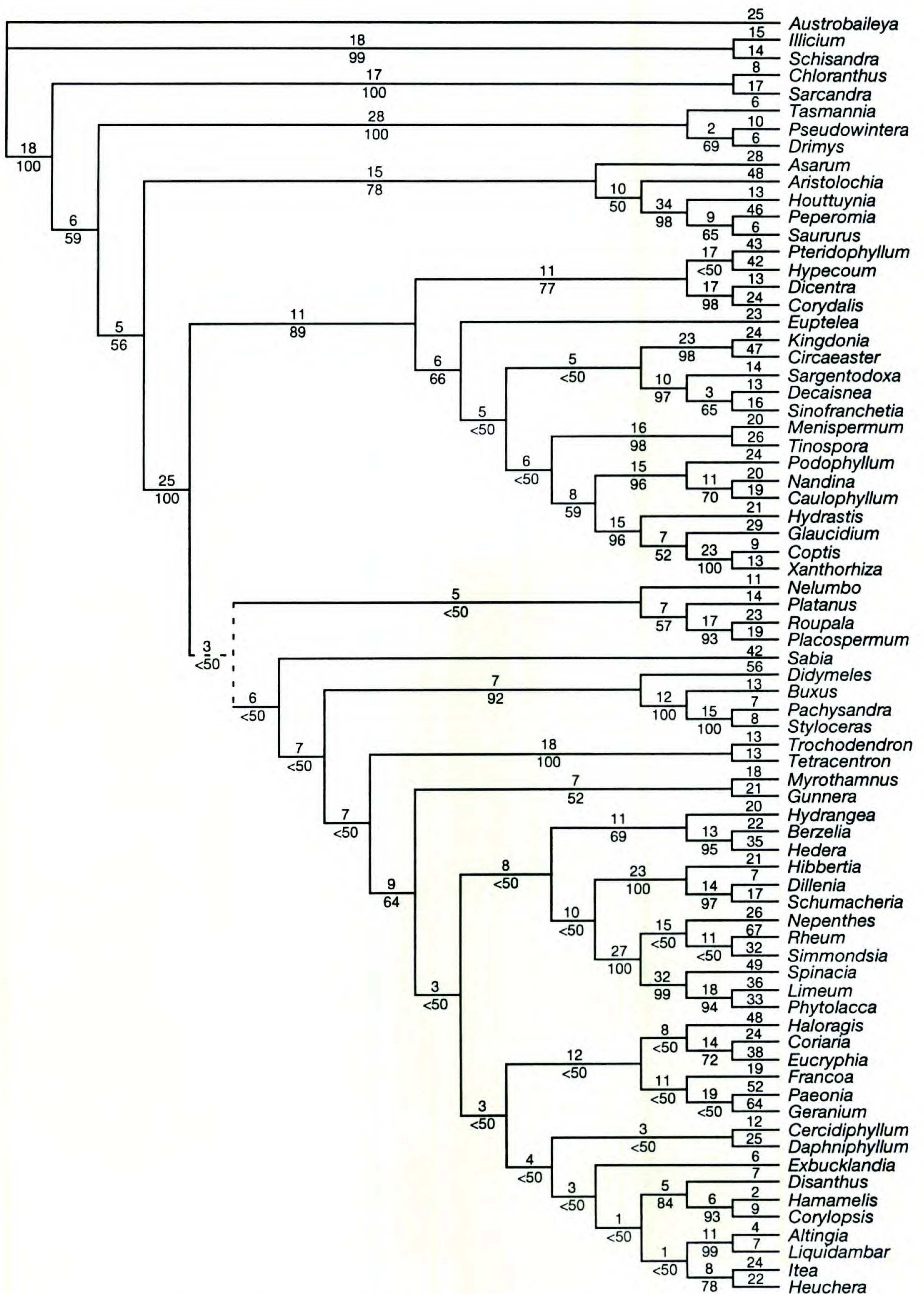


Figure 2. One of two shortest trees based on the *rbcL* sequence data. Numbers above and below the lines and dotted lines are as in Figure 1. See Table 3 for tree statistics.

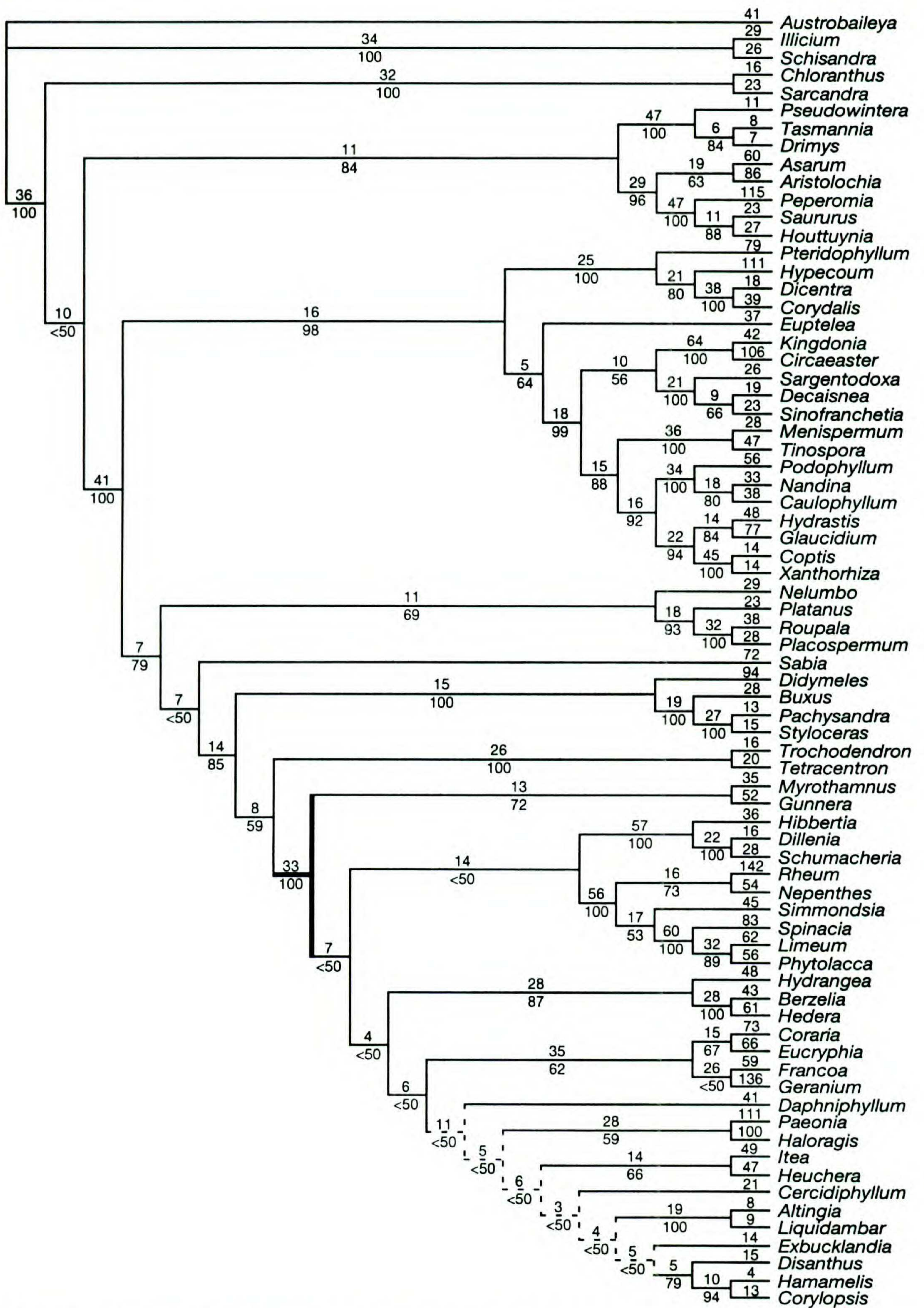


Figure 3. One of six shortest trees based on a combination of *atpB* and *rbcL* sequence data. Numbers above and below the lines and dotted lines are as in Figure 1. Node in bold indicates the well-supported core eudicot clade. See Table 3 for tree statistics.

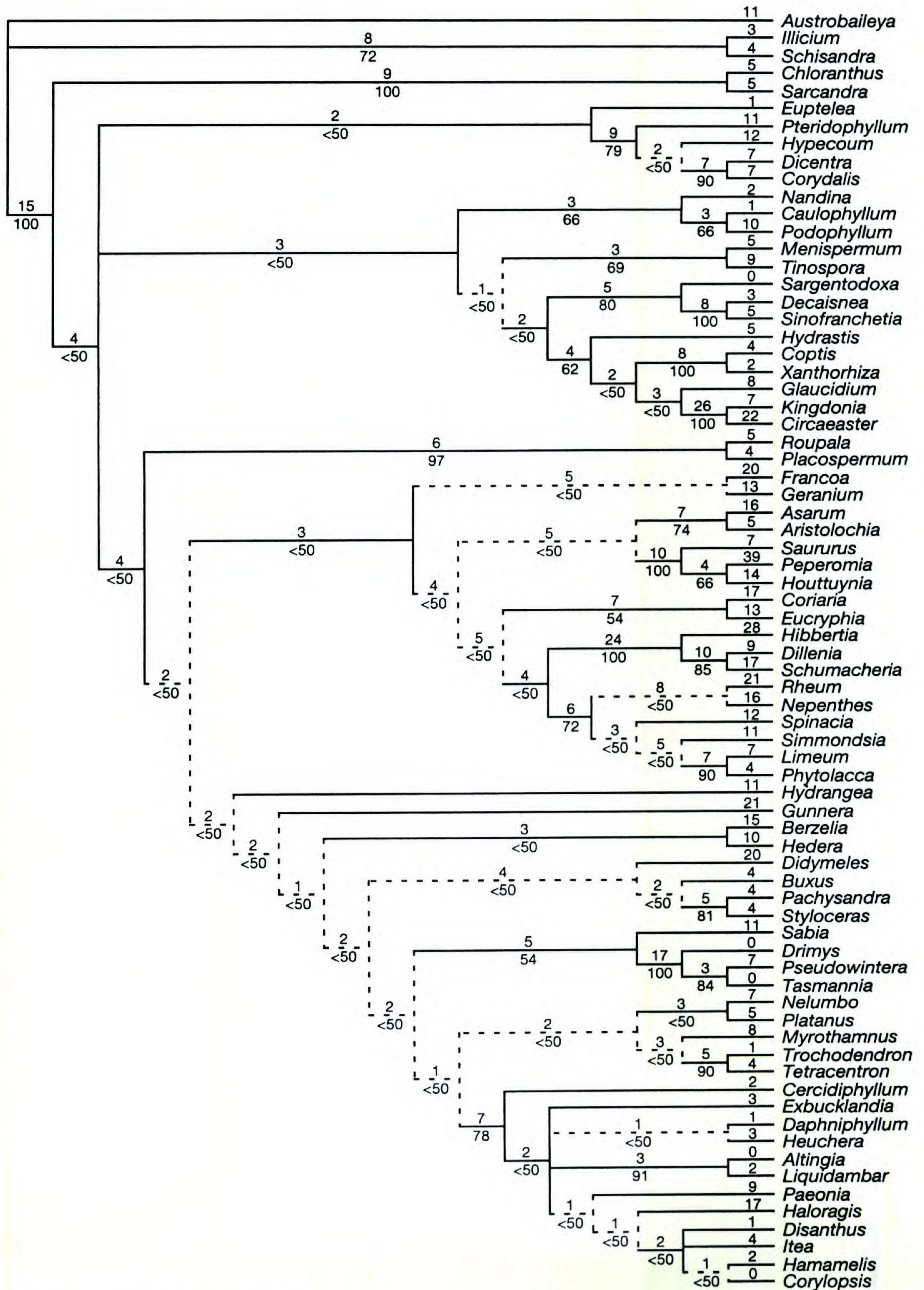


Figure 4. One of 270 shortest trees based on the nr18S sequence data. Numbers above and below the lines and dotted lines are as in Figure 1. See Table 3 for tree statistics.

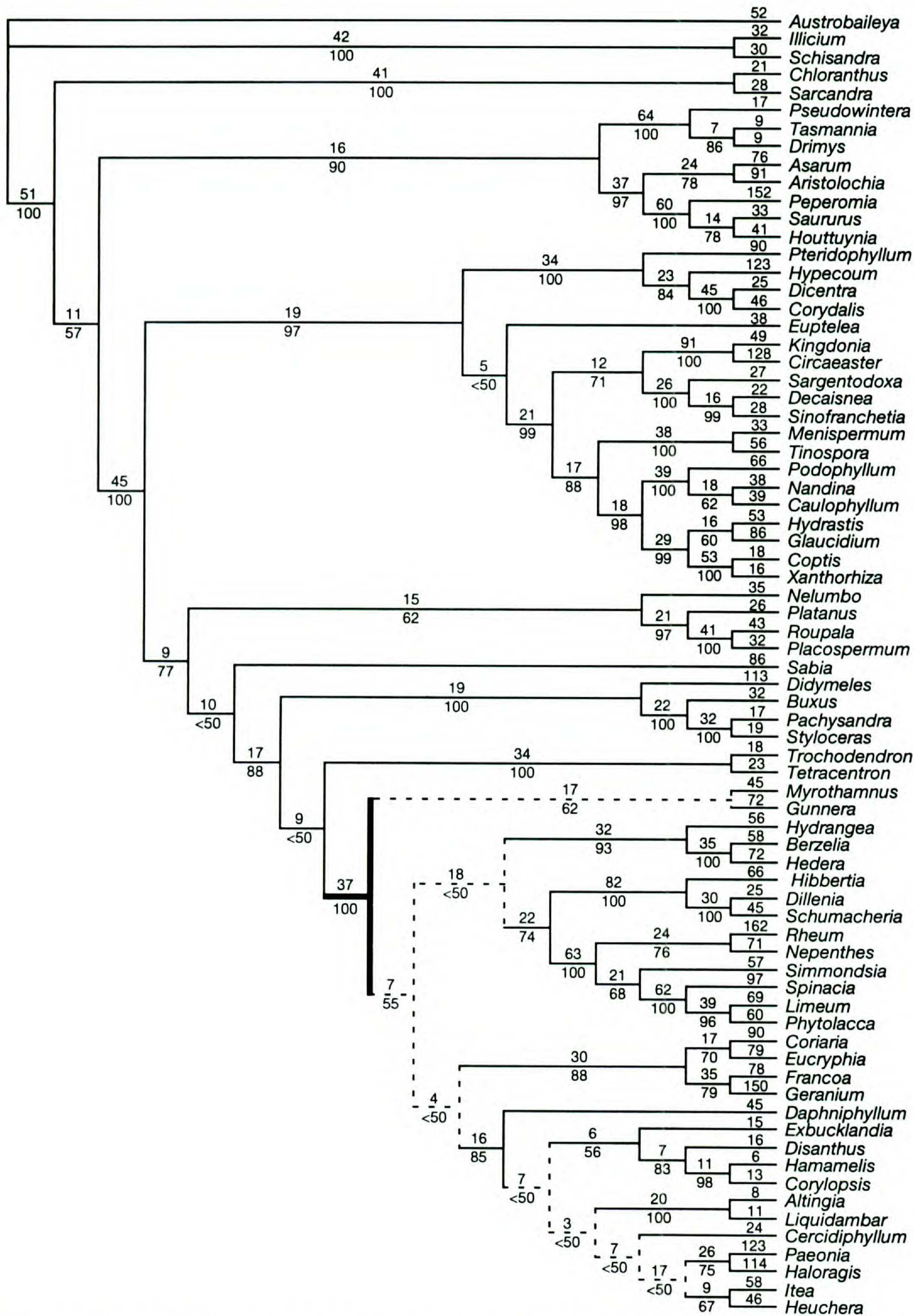


Figure 5. One of 15 shortest trees based on a combination of *atpB*, *rbcL*, and 18S sequence data. Numbers above and below the lines and dotted lines are as in Figure 1. Node in bold indicates the well-supported core eudicot clade. See Table 3 for tree statistics.

results of the combined chloroplast data analysis will be discussed here in detail (Fig. 3).

The six trees resulting from analysis of the combined chloroplast data (*atpB* and *rbcL*) strongly support the monophyly of the eudicots as a whole (bootstrap = 98%). Within the group there are two major well-supported clades, the Ranunculidae (including *Euptelea*) and the “core eudicots” (Fig. 3, node highlighted in bold).

In Ranunculidae, there is strong support for the monophyly of traditionally recognized taxa including Papaverales, Circaeasteraceae (including *Kingdonia*), Lardizabalaceae (including *Sargentodoxa*), Menispermaceae, Berberidales, and Ranunculaceae. Within Ranunculidae, Papaverales and *Euptelea* are basal, while Berberidales and Ranunculaceae are found in a derived position.

Within the non-ranunculid “lower” eudicots, there is moderate support for an expanded hamamelid clade (bootstrap = 79%) consisting of several basal eudicot lineages forming a paraphyletic series: (1) *Platanus*/Proteaceae/*Nelumbo*, (2) *Sabia*, (3) Buxaceae/*Didymeles*, (4) *Trochodendron*/*Tetracentron*, and (5) the core eudicots. The core eudicots are well supported (33 substitutions, bootstrap = 100%) and consist of a highly unresolved clade of “lower” hamamelids and placeholders for various eudicot groups (Fig. 3). The “lower” hamamelids, as previously suggested, are clearly polyphyletic (Crane, 1989; Albert et al., 1994; Chase et al., 1993; Drinnan et al., 1994; Williams et al., 1994; Soltis et al., 1997; Qiu et al., 1998).

RESULTS BASED ON 18S SEQUENCE DATA

The consensus tree resulting from 18S nuclear ribosomal DNA (Fig. 4) is less resolved, and deeper branching patterns are less well supported (lower bootstrap values), than in the consensus trees resulting from the individual chloroplast data sets. This is highlighted in Table 5, which contrasts the level of support for the chloroplast and 18S data at the order/subclass level. Major factors contributing to this lack of basal support in the analyses based on 18S data are: less information (approximately half as many variable and informative sites compared to *atpB* and *rbcL*, Table 3) and a larger number of sites in highly variable regions that exhibit high levels of homoplasy (Hoot, unpublished data). At lower taxonomic levels, however, many of the more terminal clades with adequate bootstrap support are identical to those found in the cladograms based on the chloroplast sequence data (Table 6).

The anomalous placement of some magnoliid taxa within the eudicots seen in previous analyses

of 18S data (Nickrent & Soltis, 1995; Soltis et al., 1997) is also found in the analysis presented here (Fig. 4). Interestingly, although the sampling differs in the two studies, two of the same families are involved: Aristolochiaceae and Winteraceae. In our 18S analysis, the paleoherb order Piperales is also placed within the eudicots, while it remained nested among other magnoliid groups in the Soltis et al. (1997) analysis. However, in both analyses, the branches involved in this anomalous placement of magnoliid taxa have bootstrap values < 50%. Indeed, if all branches with bootstrap values < 50% are collapsed, the tree (Fig. 4) reads as a highly unresolved polytomy with little or no information about higher-level taxonomic relationships.

RESULTS BASED ON COMBINED *RBCL*, *ATPB*, AND 18S SEQUENCE DATA

The trees (Figs. 5, 6) based on a combination of all the data are very similar in topology to the trees derived from analyses of the data from the two chloroplast genes, but, in general, the addition of 18S data resulted in higher bootstrap support for many of the branches (Tables 5, 6). Four clades are not supported (bootstrap values \leq 50%) by any of the individual data sets, but are supported in the combined analyses (Tables 5, 6): Circaeasteraceae/Lardizabalaceae, Rosid I & II, *Nelumbo*/*Platanus*/Proteaceae, and Hamamelidaceae s. str. (i.e., Hamamelidoideae sensu Endress, 1989a, 1993).

The results of the first four alternative analyses with varied taxon sampling (see “Materials and Methods”) were very similar in tree topology to the tree resulting from complete sampling (Fig. 5). Varying the outgroup by removing either the paleoherbs or the “woody” magnoliids resulted in trees with the same topology as presented in Figure 5, except that *Euptelea* formed a trichotomy within the ranunculids when only woody magnoliids were used as the outgroup, whereas it was resolved as sister group to all ranunculids except Papaverales when the paleoherbs were the outgroup. Reducing the sample size of the ranunculids to three or six genera (see “Materials and Methods”) resulted in identical tree topologies to the tree based on complete sampling with only one exception: the tree based on three ranunculid genera resulted in a lack of resolution for *Sabia* in relation to other eudicots in the “lower” hamamelid/core eudicot clade (Fig. 6). Analysis of just the core eudicot group (as delimited in Fig. 6) using *Tetracentron* as an outgroup, resulted in one fully resolved tree. However, bootstrap support was < 50% for the identical branches

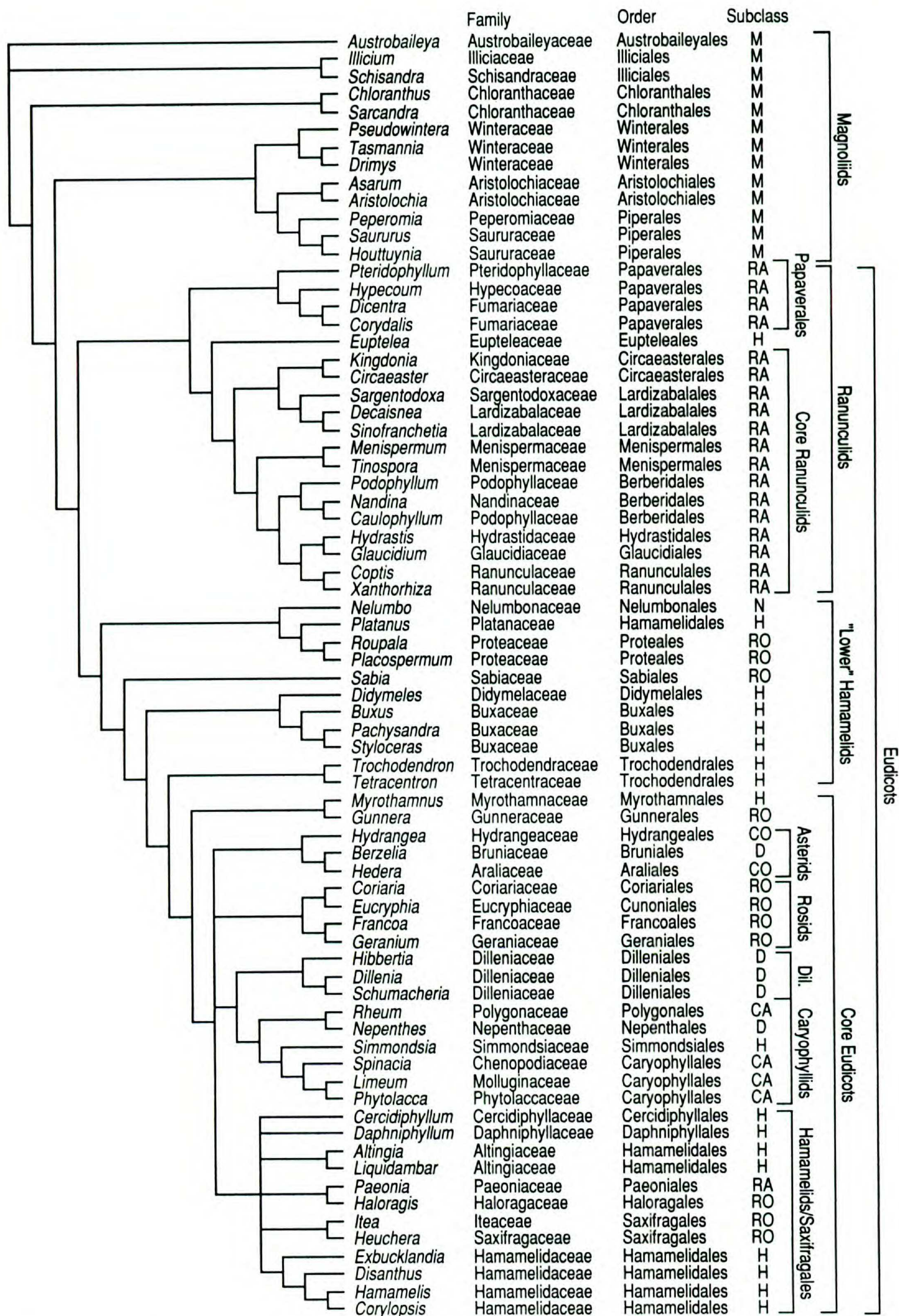


Figure 6. Strict consensus tree of the 15 shortest trees obtained from the combined data (*atpB*, *rbcL*, and 18S). Families, orders, and subclass acronyms are listed in the first three columns (taxonomic designations according to

that collapsed in the strict consensus tree derived with complete taxon sampling (dotted lines, Fig. 5).

DISCUSSION

As in other work involving the combination of large molecular data sets (e.g., Hoot et al., 1995, 1997; Soltis et al., 1998), the analyses based on the combined data (both the combined chloroplast sequences and a combination of all three sequences) had shorter computer run times and resulted in trees with better resolution and improved bootstrap support. The increases in resolution and support for groupings at the subclass and family levels are indicated in Tables 5 and 6. Of the individual data sets, bootstrap values and resolution were the highest with *atpB* and *rbcL* sequences from the chloroplast genome and the lowest with nr18S DNA sequences. In addition, the tree topology derived from a combination of all the data was not sensitive to variations in either outgroup or ingroup sampling. Any differences occurring as a result of various sampling experiments were minor and confined to branching patterns with weak support.

In the following paragraphs, we briefly discuss the congruence of our results with previous work based on non-molecular characters, with an emphasis on floral morphology. A more detailed morphological study of the "lower" eudicots is in progress (Magallón & Crane).

RANUNCULID CLADE

A ranunculid clade, which corresponds largely to subclass Ranunculidae of Takhtajan (1997), is strongly supported (bootstrap = 97%, 19 substitutions; Fig. 5) in the combined analysis of *atpB*, *rbcL*, and 18S sequence data. The ranunculids are characterized by unusually large and homogeneous S₁ sieve element plastids (Behnke, 1995), benzylisoquinoline alkaloids of the berberine and morphine type (Jensen, 1995), and epicuticular wax tubules (also found in a few non-ranunculid families, including Nelumbonaceae; Barthlott & Theisen, 1995). There are no clear synapomorphies in floral morphology for the Ranunculidae (Endress, 1995). The flowers are often large and conspicuous with brightly colored petals with whorled phyllotaxis, but there are many exceptions. A di- or trimerous

floral plan seems to be basic in the group (Drinnan et al., 1994).

Euptelea. A surprising result that emerged from previous studies (Chase et al., 1993; Drinnan et al., 1994; Hoot & Crane, 1995; Soltis et al., 1997), and that is supported by this work utilizing three gene sequences and more extensive sampling, is the inclusion of *Euptelea* within the ranunculid clade in a basal position near the Papaverales. Because of the low bootstrap value (< 50%) in the total evidence tree, phylogenetic patterns involving *Euptelea* are best considered as a trichotomy that includes Papaverales and the remaining "core" ranunculids (Figs. 5, 6).

Euptelea has traditionally been placed among the "lower" hamamelids, characterized by relatively inconspicuous flowers (e.g., *Trochodendron*, *Cercidiphyllum*; Endress, 1986; Cronquist, 1981; Takhtajan, 1997). Flowers of *Euptelea* are small, bisexual, lack a perianth, and have a variable number of stamens and carpels, apparently in whorls. Developmental studies indicate that the floral primordium is bilaterally symmetrical (Endress, 1986), perhaps indicating a basically dimerous floral plan. The floral features of *Euptelea* are significantly different from those of other Ranunculidae. One possible similarity between the flowers of *Euptelea* and several Ranunculales and Papaverales is a dimerous floral organization, but the distribution of this character state among ranunculids and "lower" hamamelids is complex. There is apparently no uniquely derived floral feature that links this genus with Papaverales and Ranunculales.

Papaverales. A clade corresponding to the order Papaverales of Cronquist (1981) and Takhtajan (1997), represented in this study by *Pteridophyllum*, *Hypocoum*, and two genera of Fumariaceae, is solidly supported as a monophyletic group (bootstrap = 100%, 34 substitutions) and appears as sister to all remaining ranunculids (core ranunculids) in the strict consensus tree resulting from the combined three-gene analysis (Figs. 5, 6). The position of Papaverales as sister to *Euptelea* and the core ranunculids is consistent with the results of previous analyses based on molecular data (Chase et al., 1993; Hoot & Crane, 1995; Soltis et al., 1997), but conflicts with previous cladistic analyses

←

Takhtajan, 1997). Acronyms for subclasses: CA = Caryophyllidae, CO = Cornidae, D = Dilleniidae, H = Hamamelididae, M = Magnoliidae, N = Nelumbonidae, RA = Ranunculidae, RO = Rosidae. Dil. = Dilleniaceae. Brackets to the right indicate major taxonomic groups discussed in text.

Table 5. Bootstrap support (%) for various groups of order to subclass rank sensu Takhtajan (1997) found in separate and combined data analyses (Figs. 1–6). See Figure 6 for generic makeup of various groups. * As defined in Chase et al. (1993). “—” indicates bootstrap values $\leq 50\%$.

Higher taxonomic groupings	<i>atpB</i>	<i>rbcL</i>	<i>atpB/rbcL</i>	18S	<i>atpB/rbcL</i> / 18S
Winteraceae/Aristolochiaceae/Piperales	90	—	84	—	90
Piperales	98	98	100	100	100
Eudicots + <i>Nelumbo</i>	98	100	100	—	100
Ranunculidae + <i>Euptelea</i>	64	89	98	—	97
Papaverales	89	77	100	79	100
Ranunculales	95	—	99	—	99
Circaeasteraceae/Lardizabalaceae s.l.	—	—	56	—	71
Menispermaceae/Berberidaceae/Ranunculaceae s.l.	77	—	88	—	88
Berberidaceae/Ranunculaceae s.l.	85	59	92	—	98
Lower eudicots excluding ranunculids	64	—	79	—	77
Hamamelididae	—	—	—	—	—
Core eudicots (hamamelids, caryophyllids, asterids, rosids)	100	64	100	—	100
Dilleniidae	100	100	100	100	100
Rosid I & II*	—	—	62	—	88
Asterids*	52	69	87	—	93
Caryophyllids + <i>Simmondsia</i>	100	100	100	72	100
Hamamelidales + <i>Paeonia</i> , <i>Haloragis</i> , <i>Itea</i> , and <i>Heuchera</i>	66	—	—	78	85

of morphological data (Loconte & Stevenson, 1991; Loconte et al., 1995), which placed Papaverales as sister to Ranunculaceae in a relatively derived position with respect to other ranunculid families.

Based on results from the combined *atpB*, *rbcL*, and 18S data, the latter conclusion is unparsimonious. Moving Papaverales into a derived position as sister to Ranunculaceae adds an additional 24 steps (total

Table 6. Bootstrap support (%) for various clades at the family level. “—” indicates bootstrap values $\leq 50\%$.

Family-level groupings	<i>atpB</i>	<i>rbcL</i>	<i>atpB/rbcL</i>	18S	<i>atpB/rbcL</i> / 18S
<i>Illicium/Schisandra</i>	97	99	100	72	100
Chloranthaceae	100	100	100	100	100
Winteraceae	100	100	100	100	100
Aristolochiaceae	65	—	63	74	78
Saururaceae	93	—	88	—	78
Fumariaceae (including <i>Hypecoum</i>)	83	—	80	—	84
Circaeasteraceae	100	98	100	100	100
Lardizabalaceae s.l. (including <i>Sargentodoxa</i>)	100	97	100	80	100
Lardizabalaceae s. str.	54	65	66	100	99
Menispermaceae	100	98	100	69	100
Berberidaceae (including <i>Nandina</i>)	100	96	100	66	100
Ranunculaceae s.l. (including <i>Hydrastis/Glaucidium</i>)	86	96	94	62	99
Ranunculaceae s. str.	100	100	100	100	100
<i>Nelumbo/Platanus/Proteaceae</i>	—	—	69	—	62
<i>Platanus/Proteaceae</i>	96	57	93	—	97
Proteaceae	100	93	100	97	100
<i>Trochodendron/Tetracentron</i>	100	100	100	90	100
Buxaceae/Didymelaceae	89	92	100	—	100
Buxaceae	94	100	100	—	100
Hamamelidaceae s.l. (including <i>Altingia/Liquidambar</i>)	—	—	—	—	—
Hamamelidaceae s. str.	—	—	—	—	56

tree length = 5714 steps); moving Ranunculaceae into a relatively basal position as sister to Papaverales incurs an additional 25 steps.

Papaverales constitute a coherent group that has been recognized traditionally as a distinct natural entity, characterized by paracarpous gynoecia and the presence of secretory idioblasts or laticifers (except for *Pteridophyllum*; Kadereit et al., 1995; Hoot et al., 1997). The flowers are bisexual and range from regularly symmetrical to markedly zygomorphic. They have a whorled and basically dimorous, opposite-decussate organization.

Pteridophyllum is strongly supported as sister to Hypecoaceae plus Fumariaceae based on the analysis of the combined data (Fig. 5). The flowers of *Pteridophyllum* have a perianth composed of one pair of sepals and two pairs of petals. The androecium consists of four stamens arranged in two opposite-decussate pairs, and the gynoecium has two connate carpels, forming a unilocular ovary.

The sister-group relationship between *Hypecoum* and Fumariaceae is well supported (bootstrap = 84%, 23 substitutions; Fig. 5). The flowers of *Hypecoum* have a similar organization to those of *Pteridophyllum* but differ in that the stamens of the inner pair have anthers in which the two thecae are separated. The gynoecium is unilocular, formed by two connate carpels.

Results of the combined three-gene analysis indicate that the two representatives of Fumariaceae s. str. (*Dicentra*, *Corydalis*) are monophyletic (bootstrap = 100%, 45 substitutions; Fig. 5). The flowers of Fumariaceae are strongly zygomorphic, which is clearly a synapomorphy with respect to the condition found in *Pteridophyllum* and *Hypecoum*. The androecium consists of two compound stamens, each with a central bithecal portion and two lateral monothechal portions. The gynoecium has two connate carpels that form a unilocular ovary. Flowers of Papaveraceae, which were not sampled in this study, are constructed upon the same basic pattern as those of *Pteridophyllum*, *Hypecoum*, and Fumariaceae. In some Papaveraceae, the production of numerous stamens and the development of multiple internal placentae in unilocular ovaries formed from one to a few carpels are clearly derived features (Hoot et al., 1997).

"CORE" RANUNCULIDS

A "core" ranunculid clade composed of Circaeasteraceae, Lardizabalaceae, Menispermaceae, Hydrastidaceae, Glaucidiaceae, Berberidaceae, and Ranunculaceae is strongly supported (bootstrap = 99%, 21 substitutions) in the combined analysis

of *atpB*, *rbcL*, and 18S sequence data (Figs. 5, 6). This clade corresponds approximately to the order Ranunculales of Cronquist (1981). Floral morphology in the core ranunculid group is extremely varied. Flowers may be unisexual (e.g., Lardizabalaceae, Menispermaceae) or bisexual, with radial to bilateral symmetry. Merosity varies from dimerous (e.g., *Circaeaster*, *Glaucidium*) or trimerous (e.g., Lardizabalaceae, Menispermaceae, Berberidaceae) to 4–12-merous with helical or whorled phyllotaxy (e.g., Ranunculaceae). Hypogyny and apocarpous are characteristic of this group.

Circaeasteraceae s.l. *Circaeaster* and *Kingdonia* form a moderately well-supported sister group to Lardizabalaceae and *Sargentodoxa* (bootstrap = 71%, 12 substitutions) in the tree resulting from a combination of all the data (Fig. 5). Morphological comparisons of Circaeasteraceae with *Sargentodoxa* and Lardizabalaceae s. str. (e.g., *Decaisnea* and *Sinofranchetia*) reveal little similarity in vegetative and floral features. Significant differences between Circaeasteraceae s.l. and Lardizabalaceae s.l. are also reflected in the unusually long branches associated with Circaeasteraceae and its two genera, *Circaeaster* and *Kingdonia* (Figs. 1–5).

Circaeaster and *Kingdonia* form an extremely well-supported monophyletic group (bootstrap = 100%, 91 substitutions; Fig. 5). This is consistent with past interpretations (Foster, 1961, 1963; Oxelman & Liden, 1995), but conflicts with suggestions that *Kingdonia* should be placed within Ranunculaceae in a position close to *Anemone* (Tamura, 1962, 1995; Kosuge et al., 1989). Based on our combined data set, moving *Kingdonia* to the base of Ranunculaceae s.l. (represented here by four genera) adds 82 steps to the shortest tree length. Experiments involving a larger sampling of Ranunculaceae are similarly unparsimonious (Hoot, unpublished results).

In terms of floral organization and life history, *Circaeaster* and *Kingdonia* differ markedly. The flowers of *Circaeaster* are usually bisexual, with a whorled phyllotaxy and irregular floral merosity. Most flowers have a perianth of two or three undifferentiated tepals, two stamens, and one carpel (Hu et al., 1990). The flowers of *Kingdonia* are bisexual, also with irregular and variable floral merosity. Flowers are apparently based on a helical phyllotaxy (Kosuge et al., 1989; Drinnan et al., 1994). There are 5 to 7 undifferentiated tepals, 8 to 12 external staminodes, 3 to 6 internal stamens, and 5 to 8 helically arranged carpels. In addition, *Circaeaster* is an annual and *Kingdonia* is a perennial, rhizomatous plant. However, the dichotomous venation in the leaves of both genera is one clear and

remarkable morphological character that unites *Circaeaster* and *Kingdonia*.

Lardizabalaceae. The monophyly of Lardizabalaceae s. str. and the sister-group relationship of *Sargentodoxa* to Lardizabalaceae are both strongly supported (bootstrap = 99% and 100%, respectively; Figs. 5, 6). Lardizabalaceae s. str. are monoecious or dioecious. The flowers have a whorled phyllotaxy with regular symmetry. The perianth usually consists of six petaloid sepals and six petals, each arranged in two whorls. The petals are thought to be staminodial in origin (Drinnan et al., 1994). In staminate flowers, the androecium usually consists of six stamens that are opposite the petals, and pistillodes may be present. Pistillate flowers usually have three free carpels, but in some genera there may be more. Species of *Sargentodoxa* are andromonoecious or dioecious and have flowers with 48–90 helically arranged, free carpels (Cronquist, 1981; Wu & Kubitzki, 1993).

Menispermaceae. Menispermaceae is well supported as sister to a clade consisting of Berberidaceae and Ranunculaceae s.l. (bootstrap = 88%, 17 substitutions; Figs. 5, 6) and as a monophyletic family (bootstrap = 100%, 38 substitutions). Menispermaceae share a trimerous floral construction with most Berberidaceae but differ in being dioecious. The perianth is formed by relatively differentiated sepals and petals, organized in two whorls of three sepals and two whorls of three petals. The staminate flowers have six stamens in two whorls, and the pistillate flowers have mostly three (occasionally one to six) free carpels in one or more whorls, forming an apocarpous gynoecium.

Berberidaceae. The sister-group relationship between Berberidaceae and Ranunculaceae s.l. (including *Hydrastis* and *Glaucidium*) is stable and strongly supported (bootstrap = 98%, 18 substitutions; Fig. 5). This relationship is also supported by an important phytochemical character, the presence of the isoquinoline alkaloid berberine in Berberidaceae and the basal members of Ranunculaceae s.l. (Hegnauer, 1966; Jensen, 1995). Molecular data provide no support for a sister-group relationship between Berberidaceae and Papaveraceae proposed on the basis of several shared morphological characters (i.e., gynoecium congenitally closed to the level of the stigma, placentae protruding-diffuse in some genera, fruit dehiscence transverse; Endress, 1995).

The placeholders for Berberidaceae form a strongly supported monophyletic group in the analyses presented here (bootstrap = 100%, 39 sub-

stitutions; Figs. 5, 6). The typically trimerous flowers of Berberidaceae are bisexual with regular symmetry and whorled phyllotaxy. The perianth consists of one or more whorls of three sepals, and one or more whorls of three petals, which may or may not have nectaries. Morphological differentiation between sepals and petals is not very strong. There are usually six stamens, apparently in two whorls of three, and opposite the petals. The gynoecium consists of two or three carpels, but is pseudomonomerous.

Ranunculaceae s.l. The sister-group relationship between Ranunculaceae and *Hydrastis/Glaucidium* (bootstrap = 99%, 29 substitutions; Fig. 5) derived from our data supports previous classifications and phylogenetic analyses based on morphological and molecular characters (Hoot, 1991, 1995). Given the strongly supported phylogenetic pattern among Lardizabalaceae, Menispermaceae, and Berberidaceae, it is more parsimonious to suppose a basically trimerous condition for the flowers of Ranunculaceae (including *Hydrastis* and *Glaucidium*) that has been modified within Ranunculaceae into a dimerous or pentamerous plan. The flowers of *Hydrastis* show a tendency toward trimery with a perianth of three tepals (sometimes two or four). *Glaucidium*, sister to *Hydrastis*, has bisexual, dimerous flowers with an opposite-decussate plan that consists of four tepals arranged in two pairs; numerous, spirally arranged stamens, and two partially fused carpels.

Floral morphology is diverse within Ranunculaceae, but most taxa have bisexual hypogynous flowers with regular symmetry and floral organs frequently arranged in a helical phyllotaxy on the floral receptacle. The innermost organs of the perianth are thought to be staminodial in nature (Tamura, 1965). The perianth parts may be in threes, fives, or variable numbers. The stamens and carpels are numerous, and the gynoecium is apocarpous.

"LOWER" HAMAMELID GRADE

Placed between the ranunculid clade, which constitutes a basal eudicot lineage, and the core eudicot clade, which includes the majority of the taxonomic diversity within the eudicots, is a paraphyletic grade that includes taxa of "lower" Hamamelididae and other affinities, which we term here the "lower" hamamelid grade (Fig. 6). There appear to be no universal morphological features shared among the independent lower hamamelid lineages and the core eudicots and, in spite of the limited number of lineages and species within the lower hamamelid grade, the disparity in floral form

and structure encompassed by these lineages is considerable. The flowers belonging to the lower hamamelid lineages may have a conspicuous perianth (e.g., *Nelumbo*), a perianth formed by small, bract-like organs (e.g., Buxaceae), or no perianth (e.g., *Trochodendron*). The androecium may consist of few (e.g., *Tetracentra*) to numerous (e.g., *Nelumbo*) stamens, and the gynoecium may be formed of few to many free or fused carpels. The ovaries are superior, with the exception of Trochodendrales (with the basal part of the ovary embedded in the floral receptacle). Floral merosity may be dimerous, tetramerous, pentamerous, or irregular, either by presumed reduction or increase in the number of floral organs. With the exception of Sabiaceae, the perianth is not differentiated into a single whorl of sepals and a single whorl of petals, apparently a plesiomorphic feature which also occurs in several lineages of the ranunculid clade. The generalized absence of a differentiated perianth (as described above) among basal eudicot lineages suggests that the perianth has not yet developed the specialized level of organization that characterizes the core eudicots.

Nelumbo/Platanaceae/Proteaceae. One of the main lineages of the lower hamamelid grade is a weakly supported clade (bootstrap = 62%, 15 substitutions) comprising *Nelumbo*, Proteaceae, and Platanaceae (Figs. 5, 6). The somewhat unexpected relationship of *Nelumbo* with Platanaceae/Proteaceae, and with non-magnoliid dicotyledons as a whole, has been previously suggested in other phylogenetic analyses (e.g., Chase et al., 1993; Drinnan et al., 1994). The placement of *Nelumbo* within the eudicots is supported by the observation that it produces (usually) tricolpate pollen grains (diagnostic of eudicots). Floral organization in *Nelumbo* is unique. The flowers are large, conspicuous, bisexual, and hypogynous, with irregular merosity. The perianth consists of greenish sepals arranged in opposite-decussate pairs, and numerous conspicuous petals arranged helically on the floral axis (Moseley & Uhl, 1985). The numerous stamens continue the helical phyllotactic pattern of the petals. The 2 to 30 carpels are free from one another, but embedded in a modified, terminally expanded, floral receptacle.

Proteaceae and Platanaceae are well supported as sister taxa (bootstrap = 97%, 21 substitutions; Fig. 5). There are several interesting similarities among the flowers of Proteaceae and Platanaceae, especially when considering fossil platanoids. Proteaceae have bisexual flowers with whorled phyllotaxy and a superior ovary. Except for the gynoe-

cium, floral merosity is tetramerous. The perianth consists of a single whorl of four tepals. The androecium has four stamens, each placed opposite one tepal, and the gynoecium consists of a single carpel. The flowers of *Platanus* are unisexual, individually inconspicuous, and grouped in compact, strictly unisexual, spherical inflorescences. Although sepals and petals have been described, morphological differentiation between the two types of organs is ambiguous. Staminate flowers typically have four stamens. Pistillate flowers have a variable number of reproductive organs. Several fossils attributed to Platanaceae on the basis of a distinctive combination of characters have flowers with relatively conspicuous perianth parts and fixed tetramery (Magallón-Puebla et al., 1997) or pentamery (e.g., Manchester, 1986; Friis et al., 1988; Crane et al., 1993). Developmental studies of the pistillate flowers of extant *Platanus* have also revealed an initially tetramerous organization (A. W. Douglas, pers. comm.). The occurrence of a basically tetramerous organization in the flowers of *Platanus*, together with the discovery of tetramerous fossil flowers apparently referable to Platanaceae, provides morphological links with Proteaceae, suggesting the possibility that this condition was shared by these two families and has subsequently been modified one to several times within Platanaceae.

Sabiaceae. While the inclusion of the genus *Sabia* in the basal eudicot grade has been supported by this work and previous phylogenetic analyses (e.g., Chase et al., 1993; Drinnan et al., 1994), its exact placement with respect to other basal eudicot lineages is not securely resolved. The sister-group relationship of *Sabia* and all remaining eudicots is weakly supported in the trees derived from the combined data (bootstrap < 50%, 10 substitutions; Fig. 5). The flowers of *Sabia* exhibit several seemingly derived features, and those of *Meliosma*, included in Sabiaceae (e.g., Cronquist, 1981) or segregated into its own family within Sabiales (Takhtajan, 1997), display several modifications presumably derived from the pattern in *Sabia*. Flowers of *Sabia* are bisexual (or rarely unisexual), hypogynous, pentamerous, with whorled phyllotaxy and regular symmetry. The perianth has one whorl of apparently bract-like sepals, and one whorl of relatively elongate petals. The androecium consists of one whorl of five stamens, and a 4- or 5-lobed disk between the androecium and corolla has been reported (Li, 1993). The gynoecium consists of two to three carpels (Li, 1993). The organs in the two perianth whorls and in the androecium are opposite one another. Flowers of *Meliosma* exhibit a modi-

fied pentamerous plan that resembles trimery. Sabiaceae are the only family among the basal eudicot lineages to have flowers with a perianth consisting of a single whorl of sepals and a single whorl of petals, organized under a pentamerous plan. Because of these seemingly advanced features, which appear equivalent to the condition found in the flowers of core eudicots (see below), a clear understanding of the phylogenetic placement of Sabiaceae with respect to other basal eudicot lineages (and especially the core eudicot lineages) becomes critical to understanding the pattern of floral evolution in core eudicots.

Didymelaceae/Buxaceae. The genus *Didymeles*, endemic to Madagascar, is confirmed as a well-supported sister group to Buxaceae (bootstrap = 100%; Figs. 5, 6). A close relationship between *Didymeles* and Buxaceae had been proposed previously based on morphological (e.g., Takhtajan, 1997) and molecular characters (Qiu et al., 1998). *Didymeles* is dioecious; the staminate flowers are reported as being dimerous, lacking a perianth, and with two opposite stamens. The pistillate flowers are described as unicarpellate, lacking a perianth or with bract-like tepals, and placed in opposite pairs or in groups of three (Cronquist, 1981; Takhtajan, 1997). Preliminary observations of the pistillate flowers of *D. madagascarensis* Willd. show that the pistillate flowers consist of opposite bracts or bract-like tepals at the tip of a pedicel. There is usually one large carpel subtended by one of the tepals; the opposite tepal is empty. However, in several specimens, each of the tepals subtends a carpel, one smaller than the other. The dimerous organization of the staminate flowers of *Didymeles*, and the general aspect of the carpels, suggest similarities with Buxaceae. More detailed comparisons are currently in progress (Magallón, in prep.).

Buxaceae are strongly supported as monophyletic in the results of the combined analysis of the three data sets. Within Buxaceae, the sister-group relationship between *Pachysandra* and *Styloceras*, and the placement of *Buxus* as sister to this clade are both strongly supported (bootstrap = 100%, 32 substitutions and bootstrap = 100%, 22 substitutions, respectively; Fig. 5). The inflorescences of Buxaceae usually include both staminate and pistillate flowers. The staminate flowers are organized in a dimerous, opposite-decussate plan. The perianth is weakly differentiated from the inflorescence bracts, and two or three pairs of tepals are present. The androecium is composed of two or three pairs of stamens, and the staminate flowers contain a central pistillode. The pistillate flowers are difficult

to delimit because there is no clear-cut morphological distinction between the opposite-decussate pairs of inflorescence bracts and the helically arranged tepals. Typically there are three (rarely two) carpels united in a syncarpous ovary.

Trochodendrales. Buxaceae and *Didymeles* form a well-supported sister group to Trochodendrales and the core eudicots (bootstrap = 88%, 17 substitutions; Fig. 5). However, the relationship of Trochodendrales as sister group to the core eudicots is not supported (bootstrap < 50%, 9 substitutions), and it seems best to consider the phylogenetic placement of Trochodendrales as yet unresolved within the lower hamamelid grade. However, a sister-group relationship between *Trochodendron* and *Tetracentron* is solidly supported in the combined three-gene analysis (bootstrap = 100%, 34 substitutions; Fig. 5).

Although superficially different, the flowers of *Trochodendron* and *Tetracentron* share several distinctive attributes. Both genera have bisexual flowers with semi-inferior ovaries. The flowers of *Tetracentron* are clearly constructed on a dimerous plan with four tepals and four stamens arranged in opposite-decussate pairs. The gynoecium consists of four carpels, basally embedded in the floral receptacle and placed diagonally with respect to the tepal and stamen pairs. The flowers of *Trochodendron* are different from those of *Tetracentron*, but there is some indication that they are derived from an originally dimerous plan. Early in floral development, the floral meristem is bilaterally symmetrical. The perianth is absent in adult flowers, but two small, irregularly placed scales between the prophylls and the androecium have been interpreted as possible remains of a reduced perianth (Endress, 1986). The androecium is composed of numerous, irregularly arranged stamens. The gynoecium consists of several to many (4 to 17) collaterally arranged carpels which are embedded proximally in the floral receptacle. The pattern of carpellary fusion, the mode of fruit dehiscence, and especially, the presence of a nectarial surface on the abaxial surface of the carpels and the absence of vessels in the wood (Endress, 1986), are some of the features shared by these two genera.

"CORE" EUDICOTS

The strong support for the "core" eudicot clade (including *Myrothamnus* and *Gunnera*; bootstrap = 100%, 37 substitutions) is one of the most significant results from our combined analysis of *atpB*, *rbcL*, and 18S sequence data (Figs. 5, 6). A similar clade was found in other phylogenetic analyses

based on one gene and/or with less thorough sampling of the basal eudicot taxa (e.g., Olmstead et al., 1992; Chase et al., 1993; Drinnan et al., 1994; Williams et al., 1994; Soltis et al., 1997). Except for the inclusion of *Myrothamnus* (not sampled in previous studies) and *Gunnera*, our "core" eudicot clade is comparable to the "higher eudicots" recognized by Chase et al. (1993).

The strict consensus tree obtained from the combined analysis of the three-gene data sets (Fig. 6) recognizes a major polytomy of the following relatively well-supported clades within the core eudicots: (1) an asterid clade consisting of placeholders *Hydrangea*, *Berzelia*, and *Hedera*; (2) a clade equivalent to "rosid I and II" in search II of Chase et al. (1993), consisting of the four relatively derived placeholders *Francoa*, *Geranium*, *Coriaria*, and *Eucryphia*; (3) a caryophyllid clade plus Dilleniaceae (*Rheum*, *Nepenthes*, *Simmondsia*, *Spinacia*, *Limeum*, *Phytolacca*, *Hibbertia*, *Dillenia*, and *Schumacheria*); and (4) a hamamelid/Saxifragales clade (*Cercidiphyllum*, *Daphniphyllum*, *Altingia*, *Liquidambar*, *Paeonia*, *Haloragis*, *Itea*, *Heuchera*, *Exbucklandia*, *Disanthus*, *Hamamelis*, and *Corylopsis*) comparable to the rosid III clade of Chase et al. (1993) and the "saxifragoids" of Soltis et al. (1997). The poorly supported and unstable phylogenetic patterns among the major clades of the core eudicots (Figs. 5, 6), together with the conflicting patterns of relationships obtained in independent analyses (e.g., Chase et al., 1993; Morgan & Soltis, 1993; Williams et al., 1994; Soltis & Soltis, 1997), suggest that relationships among these major clades are currently best expressed as a polytomy.

Floral morphology among the core eudicots is extremely diverse. However, the core eudicot floral groundplan apparently consists of a whorled phyllotaxy with a fixed number of organs in each whorl (usually five in calyx and corolla, five or ten in the androecium, and two in the gynoecium) and alternation in the radial placement of organs of adjacent floral whorls. The perianth consists of a single whorl of sepals and a single whorl of petals that are morphologically differentiated from each other.

Myrothamnus/Gunnera. The sister-group relationship of *Myrothamnus* and *Gunnera* is not well supported (bootstrap = 62%, 17 substitutions; Fig. 5) and these two genera are best thought of as forming an unresolved polytomy with clades that include the remaining core eudicots. These two genera differ greatly in vegetative and reproductive morphology. *Myrothamnus* is a shrub with small, xeromorphic, fan-like leaves. The plants are dioecious, and the flowers have a perianth generally

formed by four bract-like tepals. The androecium consists of four stamens, apparently arranged in a single whorl. The pistillate flowers have three or four carpels that are basally fused and distally free, and are reported to alternate with the tepals (Endress, 1989b; Kubitzki, 1993).

Gunnera consists of herbaceous plants that produce tiny (e.g., *G. magellanica* Lam.) to gigantic leaves (e.g., *G. chilensis* Lam.). The flowers are usually unisexual, apparently dimerous, and epigynous. The perianth is formed by two or three small sepals and two petals that are larger than the sepals. The androecium consists of one or two stamens, and the gynoecium of two fused carpels. The placement of organs with respect to those of neighboring whorls is not well understood.

Caryophyllids/Dilleniaceae. The phylogenetic placement of Dilleniaceae within the core eudicots has been problematic (e.g., Chase et al., 1993; Morgan & Soltis, 1993; Rice et al., 1997). In this study, a sister-group relationship between Dilleniaceae and the caryophyllid clade is supported (bootstrap = 74%, 22 substitutions; Figs. 5, 6). The caryophyllids and Dilleniaceae are each supported as monophyletic with high bootstrap values (100%; Fig. 5). The genus *Simmondsia*, frequently classified close to Buxaceae, although generally regarded as constituting a monotypic and somewhat isolated family (e.g., Cronquist, 1981; Takhtajan, 1997), appears within the caryophyllid clade in a weakly supported sister-group relationship with Caryophyllales (bootstrap = 68%, 21 substitutions; Fig. 5). A close relationship between *Simmondsia* and the Centrospermae (i.e., Caryophyllales) was proposed by van Tieghem (1897) and is supported by palynological, chemical, and microstructural characters (Jarvis, 1989).

Hamamelids/Saxifragales. In our analysis of the combined three-gene data sets, the "hamamelid/Saxifragales" clade is recognized as a monophyletic group with moderate support (bootstrap = 85%, 16 substitutions; Fig. 5). The occurrence of the hamamelid/Saxifragales clade as an independent lineage within the core eudicots has been reported in analyses that include a broad sample of eudicots (e.g., Rice et al., 1997). Alternatively, the hamamelids/Saxifragales have been resolved as sister to a largely rosid clade (e.g., Chase et al., 1993; Soltis et al., 1997). Most clades within the hamamelid/Saxifragales clade are poorly supported (bootstrap < 70%; Fig. 5). The only clades with reliable support are: *Disanthus/Hamamelis/Corylopsis* (Hamamelidaceae), *Altingia/Liquidambar* (Altingiaceae), and *Paeonia/Haloragis*.

CONCLUSIONS

The analyses presented here were designed to explore the pattern of phylogenetic relationships at the base of the eudicot clade. Data from *rbcL*, *atpB*, and 18S genomic sequences were analyzed independently and in combination to provide an assessment of phylogenetic patterns among an extensive representation of previously identified basal eudicot taxa, as well as selected representatives of more derived clades. Because phylogenetic patterns among major angiosperm lineages and the sister-group relationships for the eudicot clade are not yet resolved, the outgroup comprised a taxonomically broad selection of magnoliid lineages, including herbaceous and woody representatives. Analyses in which different magnoliid taxa were used as outgroups documented the stability of ingroup phylogenetic patterns.

Analyses of *rbcL*, *atpB*, combined *rbcL/atpB*, and combined *rbcL/atpB/18S* sequence data support the previously detected basic structure of the eudicot clade, with the ranunculid clade (Papaverales and Ranunculales) forming the sister group to all other eudicots ("lower" hamamelids plus core eudicots). Within the "lower" hamamelid/core eudicot clade several independent lineages, mostly of "lower" Hamamelididae, constitute a grade leading to a major clade, the core eudicots. The core eudicots include taxa of Caryophyllidae, Dilleniidae, Rosidae, and Asteridae, as well as some additional genera of "lower" Hamamelididae. The use of placeholders to represent the ranunculid clade did not have any major effect on the pattern of relationships among the remaining basal eudicots.

Among previously suggested phylogenetic patterns that were confirmed by this study are: the placement of *Euptelea* within the ranunculid clade; the close relationship of Berberidaceae to Ranunculaceae rather than Papaverales; the inclusion of *Nelumbo* within the eudicot clade and its placement among the lineages of the "lower" hamamelid grade; and the detection of major caryophyllid, asterid, and rosid clades within the core eudicot clade. A result that emerged in other phylogenetic analyses, but which is not universally supported, is the presence of a hamamelid-rosid clade (hamamelids/Saxifragales, Fig. 6) separate from more derived rosids, forming an independent branch within the core eudicot clade. The sister-group relationship of *Didymeles* with Buxaceae and the close relationship of *Simmondsia* with Caryophyllales, both of which had been suggested previously based on traditional analysis of morphological evidence, are strongly supported by our molecular results.

Although this study provides considerable information about phylogenetic patterns at the base of the eudicot clade, issues that still require resolution include: the placement of Sabiaceae among the basal eudicot lineages; whether the hamamelid/Saxifragales clade is part of the main rosid clade or constitutes an independent lineage within the core eudicots; and the monophyletic status of the Hamamelidaceae (sensu Endress, 1989a).

Literature Cited

- Albert, V. A., S. E. Williams & M. W. Chase. 1992. Carnivorous plants: Phylogeny and structural evolution. *Science* 257: 1491–1495.
- , A. A. Backlund, K. Bremer, M. W. Chase, J. R. Manhart, B. D. Mishler & K. C. Nixon. 1994. Functional constraints and *rbcL* evidence for land plant phylogeny. *Ann. Missouri Bot. Gard.* 81: 534–567.
- Angiosperm Phylogeny Group (APG). 1998. An ordinal classification for the families of flowering plants. *Ann. Missouri Bot. Gard.* 85: 531–553.
- Barthlott, W. & I. Theisen. 1995. Epicuticular wax ultrastructure and classification of Ranunculiflorae. *In* U. Jensen & J. W. Kadereit (editors), *Systematics and Evolution of the Ranunculiflorae*. *Pl. Syst. Evol. (Suppl.)* 9: 39–45.
- Behnke, H.-D. 1995. Sieve-element plastids, phloem proteins, and evolution of the Ranunculanae. *In*: U. Jensen & J. W. Kadereit (editors), *Systematics and Evolution of the Ranunculiflorae*. *Pl. Syst. Evol. (Suppl.)* 9: 25–37.
- Chase, M. W., D. E. Soltis, R. G. Olmstead, D. Morgan, D. H. Les, B. D. Mishler, M. R. Duvall, R. A. Price, H. G. Hills, Y.-L. Qiu, K. A. Kron, J. H. Rettig, E. Conti, J. D. Palmer, J. R. Manhart, K. J. Sytsma, H. J. Michaels, W. J. Kress, K. G. Karol, W. D. Clark, M. Hedrén, B. S. Gaut, R. K. Jansen, K.-J. Kim, C. F. Wimpee, J. F. Smith, G. R. Furnier, S. H. Strauss, Q.-Y. Xiang, G. M. Plunkett, P. S. Soltis, S. M. Swensen, S. E. Williams, P. A. Gadek, C. J. Quinn, L. E. Eguiarte, E. Golenberg, G. H. Learn, S. W. Graham, S. C. H. Barrett, S. Dayanandan & V. A. Albert. 1993. Phylogenetics of seed plants: An analysis of nucleotide sequences from the plastid gene *rbcL*. *Ann. Missouri Bot. Gard.* 80: 528–580.
- Cosner, M. E., R. K. Jansen & T. G. Lammers. 1994. Phylogenetic relationships in the Campanulales based on *rbcL* sequences. *Pl. Syst. Evol.* 190: 79–95.
- Crane, P. R. 1989. Paleobotanical evidence on the radiation of non-magnoliid dicotyledons. *Pl. Syst. Evol.* 162: 165–191.
- , E. M. Friis, K. R. Pedersen & A. N. Drinnan. 1993. Early Cretaceous (Early to Middle Albian) platanoid inflorescences associated with *Sapindopsis* leaves from the Potomac Group of eastern North America. *Syst. Bot.* 18: 328–344.
- Cronquist, A. 1981. *An Integrated System of Classification of Flowering Plants*. Columbia Univ. Press, New York.
- Dahlgren, R. M. T. 1980. A revised system of classification of angiosperms. *Bot. J. Linn. Soc.* 80: 90–124.
- Donoghue, M. J. & J. A. Doyle. 1989a. Phylogenetic studies of seed plants and angiosperms based on morphological characters. Pp. 181–193 *in* B. Fernholm, K. Bremer & H. Jornvall (editors), *The Hierarchy of Life:*

- Molecules and Morphology in Phylogenetic Analysis. Elsevier Science Publishers, Amsterdam.
- & ———. 1989b. Phylogenetic analysis of the angiosperms and the relationship of "Hamamelidae." Pp. 17–45 in P. R. Crane & S. Blackmore (editors), *Evolution, Systematics and Fossil History of the Hamamelidae*. Clarendon Press, Oxford.
- Doyle, J. A. & M. J. Donoghue. 1993. Phylogenies and angiosperm diversification. *Paleobiology* 19: 141–167.
- , M. J. Donoghue & E. A. Zimmer. 1994. Integration of morphological and ribosomal RNA data on the origin of angiosperms. *Ann. Missouri Bot. Gard.* 81: 419–450.
- Doyle, J. J. & J. L. Doyle. 1987. A rapid DNA isolation procedure for small quantities of fresh tissue. *Phytochem. Bull., Bot. Soc. Amer.* 19: 11–15.
- Drinnan, A. N., P. R. Crane & S. B. Hoot. 1994. Patterns of floral evolution in the early diversification of non-magnoliid dicotyledons (eudicots). *In: P. K. Endress & E. M. Friis (editors), Early Evolution of Flowers. Pl. Syst. Evol. (Suppl.)* 8: 93–122.
- Eckenrode, V. K., J. Arnold & R. B. Meagher. 1985. Comparison of the nucleotide sequence of soybean 18S rRNA with the sequences of other small subunit rRNA's. *J. Molec. Evol.* 21: 259–269.
- Endress, P. K. 1986. Floral structure, systematics and phylogeny in Trochodendrales. *Ann. Missouri Bot. Gard.* 73: 297–324.
- . 1989a. A suprageneric taxonomic classification of the Hamamelidaceae. *Taxon* 38: 371–376.
- . 1989b. The systematic position of the Myrothamnaceae. Pp. 193–200 in P. R. Crane & S. Blackmore (editors), *Evolution, Systematics, and Fossil History of the Hamamelidae*. Clarendon Press, Oxford.
- . 1993. Hamamelidaceae. Pp. 322–331 in K. Kubitzki (editor), *The Families and Genera of Vascular Plants, Vol. II. Flowering Plants. Dicotyledons* ed. K. Kubitzki, J. G. Rohweder & V. Bittrich. Springer-Verlag, Berlin.
- . 1995. Floral structure and evolution in Ranunculanae. *In: U. Jensen & J. W. Kadereit (editors), Systematics and Evolution of the Ranunculiflorae. Pl. Syst. Evol. (Suppl.)* 9: 47–61.
- Engler, A. 1930. Saxifragaceae. *In: A. Engler & K. Prantl (editors), Die natürlichen Pflanzenfamilien, Vol. 18a.* Wilhelm Engelmann, Leipzig.
- Farris, J. S., M. Källersjö, A. G. Kluge & C. Bult. 1995. Testing significance of incongruence. *Cladistics* 10: 315–319.
- Felsenstein, J. 1985. Confidence limits on phylogenies: An approach using the bootstrap. *Evolution* 39: 783–791.
- Foster, A. S. 1961. The floral morphology and relationships of *Kingdonia uniflora*. *J. Arnold Arbor.* 42: 397–416.
- . 1963. The morphology and relationships of *Circaeaster*. *J. Arnold Arbor.* 44: 299–321.
- Friis, E. M., P. R. Crane & K. R. Pedersen. 1988. Reproductive structures of Cretaceous Platanaceae. *Biol. Meddel. Kongel. Danske Vidensk. Selsk.* 31: 1–56.
- Hegnauer, R. 1966. Comparative phytochemistry of alkaloids. Pp. 211–230 in T. Swain (editor), *Comparative Phytochemistry*. Academic Press, London.
- Hillis, D. M. & J. J. Bull. 1993. An empirical test of bootstrapping as a method for assessing confidence in phylogenetic analysis. *Syst. Biol.* 42: 182–192.
- Hoot, S. B. 1991. Phylogeny of the Ranunculaceae based on epidermal microcharacters and macromorphology. *Syst. Bot.* 16: 741–755.
- . 1995. Phylogeny of the Ranunculaceae based on *atpB*, *rbcL*, and 18S nuclear ribosomal DNA sequence data. *In: U. Jensen & J. W. Kadereit (editors), Systematics and Evolution of the Ranunculiflorae. Pl. Syst. Evol. (Suppl.)* 9: 241–251.
- & P. R. Crane. 1995. Interfamilial relationships in the Ranunculidae based on molecular systematics. *In: U. Jensen & J. W. Kadereit (editors), Systematics and Evolution of the Ranunculiflorae. Pl. Syst. Evol. (Suppl.)* 9: 119–131.
- , A. Culham & P. R. Crane. 1995. The utility of *atpB* gene sequences in resolving relationships in the Lardizabalaceae, including comparisons with *rbcL* and 18S ribosomal DNA sequences. *Ann. Missouri Bot. Gard.* 82: 194–207.
- , J. W. Kadereit, F. R. Blattner, K. B. Jork, A. E. Schwarzbach & P. R. Crane. 1997. Data congruency and phylogeny of the Papaveraceae s.l. based on four data sets: *atpB* and *rbcL* sequences, *trnK* restriction sites, and morphological characters. *Syst. Bot.* 22: 575–590.
- Hu, Z.-H., J. Yang, R.-Q. Jing & Z.-M. Dong. 1990. Morphological studies of *Circaeaster agrestis*. *Cathaya* 2: 77–88.
- Jarvis, C. E. 1989. A review of the family Buxaceae Dumortier. Pp. 273–278 in P. R. Crane & S. Blackmore (editors), *Evolution, Systematics, and Fossil History of the Hamamelidae*. Clarendon Press, Oxford.
- Jensen, U. 1995. Secondary compounds of the Ranunculiflorae. *In: U. Jensen & J. W. Kadereit (editors), Systematics and Evolution of the Ranunculiflorae. Pl. Syst. Evol. (Suppl.)* 9: 85–97.
- Johansson, J. T. 1995. A revised chloroplast DNA phylogeny of the Ranunculaceae. *In: U. Jensen & J. W. Kadereit (editors), Systematics and Evolution of the Ranunculiflorae. Pl. Syst. Evol. (Suppl.)* 9: 253–261.
- & R. K. Jansen. 1993. Chloroplast DNA variation and phylogeny of the Ranunculaceae. *Pl. Syst. Evol.* 187: 29–49.
- Kadereit, J. W., F. R. Blattner, K. Jork & A. Schwarzbach. 1995. The phylogeny of the Papaveraceae s.l.: Morphological, geographical and ecological implications. *In: U. Jensen & J. W. Kadereit (editors), Systematics and Evolution of the Ranunculiflorae. Pl. Syst. Evol. (Suppl.)* 9: 133–145.
- Kim, Y.-D. & R. Jansen. 1995. Phylogenetic implications of chloroplast DNA variation in the Berberidaceae. *In: U. Jensen & J. W. Kadereit (editors), Systematics and Evolution of the Ranunculiflorae. Pl. Syst. Evol. (Suppl.)* 9: 341–349.
- Kosuge, K., F.-D. Pu & M. Tamura. 1989. Floral morphology and relationships of *Kingdonia*. *Acta Phytotax. Geobot.* 40: 61–67.
- Kubitzki, K. 1993. Myrothamnaceae. Pp. 468–469 in K. Kubitzki (editor), *The Families and Genera of Vascular Plants, Vol II. Flowering Plants. Dicotyledons*, ed. K. Kubitzki, J. G. Rohweder & V. Bittrich. Springer-Verlag, Berlin.
- Li, H.-L. 1993. Sabiaceae. Pp. 611–615 in T.-C. Huang (editor), *Flora of Taiwan, Vol. III. Angiosperms, Dicotyledons, Hamamelidaceae–Umbelliferae*. Editorial Committee of the Flora of Taiwan, Taipei.
- Loconte, H. & J. R. Estes. 1989. Phylogenetic systematics of Berberidaceae and Ranunculales (Magnoliidae). *Syst. Bot.* 14: 565–579.

- & D. W. Stevenson. 1991. Cladistics of the Magnoliidae. *Cladistics* 7: 267–296.
- , L. M. Campbell & D. W. Stevenson. 1995. Ordinal and familial relationships of ranunculid genera. *In*: U. Jensen & J. W. Kadereit (editors), *Systematics and Evolution of the Ranunculiflorae*. *Pl. Syst. Evol. (Suppl.)* 9: 99–118.
- Maddison, W. P. & D. R. Maddison. 1992. *MacClade: Interactive Analysis of Phylogeny and Character Evolution*, version 3.0. Sinauer Associates, Sunderland, Massachusetts.
- Magallón-Puebla, S., P. S. Herendeen & P. R. Crane. 1997. *Quadriplatanus georgianus* gen. et sp. nov.: Staminate and pistillate platanaceous flowers from the Late Cretaceous (Coniacian–Santonian) of Georgia, U.S.A. *Int. J. Pl. Sci.* 158: 373–394.
- Manchester, S. R. 1986. Vegetative and reproductive morphology of an extinct plane tree (Platanaceae) from the Eocene of western North America. *Bot. Gaz.* 147: 200–226.
- Manos, P. S., K. C. Nixon & J. J. Doyle. 1993. Cladistic analysis of restriction site variation within the chloroplast DNA inverted repeat region of selected Hamamelididae. *Syst. Bot.* 18: 551–562.
- Morgan, D. R. & D. E. Soltis. 1993. Phylogenetic relationships among members of Saxifragaceae sensu lato based on *rbcL* sequence data. *Ann. Missouri Bot. Gard.* 80: 631–660.
- Moseley, M. F. & N. W. Uhl. 1985. Morphological studies of the Nymphaeaceae sensu lato XV. The anatomy of the flower of *Nelumbo*. *Bot. Jahrb. Syst.* 106: 61–98.
- Nickol, M. G. 1995. Phylogeny and inflorescences of Berberidaceae—A morphological survey. *In*: U. Jensen & J. W. Kadereit (editors), *Systematics and Evolution of the Ranunculiflorae*. *Pl. Syst. Evol. (Suppl.)* 9: 327–340.
- Nickrent, D. L. & D. E. Soltis. 1995. A comparison of angiosperm phylogenies from nuclear 18S rDNA and *rbcL* sequences. *Ann. Missouri Bot. Gard.* 82: 208–234.
- Olmstead, R. G., H. J. Michaels, K. M. Scott & J. D. Palmer. 1992. Monophyly of the Asteridae and identification of their major lineages inferred from DNA sequences of *rbcL*. *Ann. Missouri Bot. Gard.* 79: 249–265.
- , B. Bremer, K. M. Scott & J. D. Palmer. 1993. A parsimony analysis of the Asteridae sensu lato based on *rbcL* sequences. *Ann. Missouri Bot. Gard.* 80: 700–722.
- Oxelman, B. & M. Liden. 1995. The position of *Circaeaster*—Evidence from nuclear ribosomal DNA. *In*: U. Jensen & J. W. Kadereit (editors), *Systematics and Evolution of the Ranunculiflorae*. *Pl. Syst. Evol. (Suppl.)* 9: 189–193.
- Plunkett, G. M., D. E. Soltis & P. S. Soltis. 1996. Higher level relationships of Apiales (Apiaceae and Araliaceae) based on phylogenetic analysis of *rbcL* sequences. *Amer. J. Bot.* 83: 499–515.
- Qui, Y.-L., M. W. Chase, S. B. Hoot, E. Conti, P. R. Crane, K. J. Sytsma & C. R. Parks. 1998. Phylogenetics of the Hamamelidae and their allies: Parsimony analyses of nucleotide sequences of the plastid gene *rbcL*. *Int. J. Pl. Sci.* 159: 881–890.
- Rice, K. A., M. J. Donoghue & R. G. Olmstead. 1997. Analyzing large data sets: *rbcL* 500 revisited. *Syst. Biol.* 46: 554–563.
- Schulze-Menz, G. K. 1964. Saxifragaceae. *In*: H. Melchior (editor), *A. Engler's Syllabus der Pflanzenfamilien*. Gebrüder Borntraeger, Berlin.
- Soltis, D. E. & P. S. Soltis. 1997. Phylogenetic relationships in Saxifragaceae sensu lato: A comparison of topologies based on 18S rDNA and *rbcL* sequences. *Amer. J. Bot.* 84: 504–522.
- , D. R. Morgan, A. Grable, P. S. Soltis & R. Kuzoff. 1993. Molecular systematics of Saxifragaceae sensu stricto. *Amer. J. Bot.* 80: 1056–1081.
- , ———, D. L. Nickrent, L. A. Johnson, W. J. Hahn, S. B. Hoot, J. A. Sweere, R. K. Kuzoff, K. A. Kron, M. W. Chase, S. M. Swenson, E. A. Zimmer, S.-M. Chaw, L. J. Gillespie, W. J. Kress & K. J. Sytsma. 1997. Angiosperm phylogeny inferred from 18S ribosomal DNA sequences. *Ann. Missouri Bot. Gard.* 84: 1–49.
- , ———, M. Mort, M. W. Chase, V. Savolainen, S. B. Hoot & C. M. Morton. 1998. Inferring complex phylogenies using parsimony: An empirical approach using three large DNA data sets for angiosperms. *Syst. Biol.* 47: 32–42.
- Takhtajan, A. 1997. *Diversity and Classification of Flowering Plants*. Columbia Univ. Press, New York.
- Tamura, M. 1962. Morphology, ecology, and phylogeny of the Ranunculaceae I. *Sci. Rep. Coll. Gen. Educ. Osaka Univ.* 14: 115–126.
- . 1965. Morphology, ecology, and phylogeny of the Ranunculaceae IV. *Sci. Rep. Coll. Gen. Educ. Osaka Univ.* 14: 53–71.
- . 1995. Phylogeny and classification of the Ranunculaceae. *In*: U. Jensen & J. W. Kadereit (editors), *Systematics and Evolution of the Ranunculiflorae*. *Pl. Syst. Evol. (Suppl.)* 9: 201–206.
- Thanikaimoni, G. 1984. *Menispermaceae: Palynologie et systématique*. *Trav. Sect. Sci. Techn., Inst. Franç. Pondichéry* 18: 1–135.
- Thorne, R. F. 1992. An updated phylogenetic classification of the flowering plants. *Aliso* 13: 365–389.
- Tieghem, P. van. 1897. Sur les Buxacées. *Ann. Sci. Nat., Bot. Sér.* 10, 5: 289–338.
- Williams, S. E., V. A. Albert & M. W. Chase. 1994. Relationships of Droseraceae: A cladistic analysis of *rbcL* sequence and morphological data. *Amer. J. Bot.* 81: 1027–1037.
- Wu, Ch.-Y. & K. Kubitzki. 1993. Lardizabalaceae. Pp. 361–365 *in* K. Kubitzki (editor), *The Families and Genera of Vascular Plants, Vol II. Flowering Plants. Dicotyledons*, ed. K. Kubitzki, J. G. Rohweder & V. Bittrich. Springer-Verlag, Berlin.